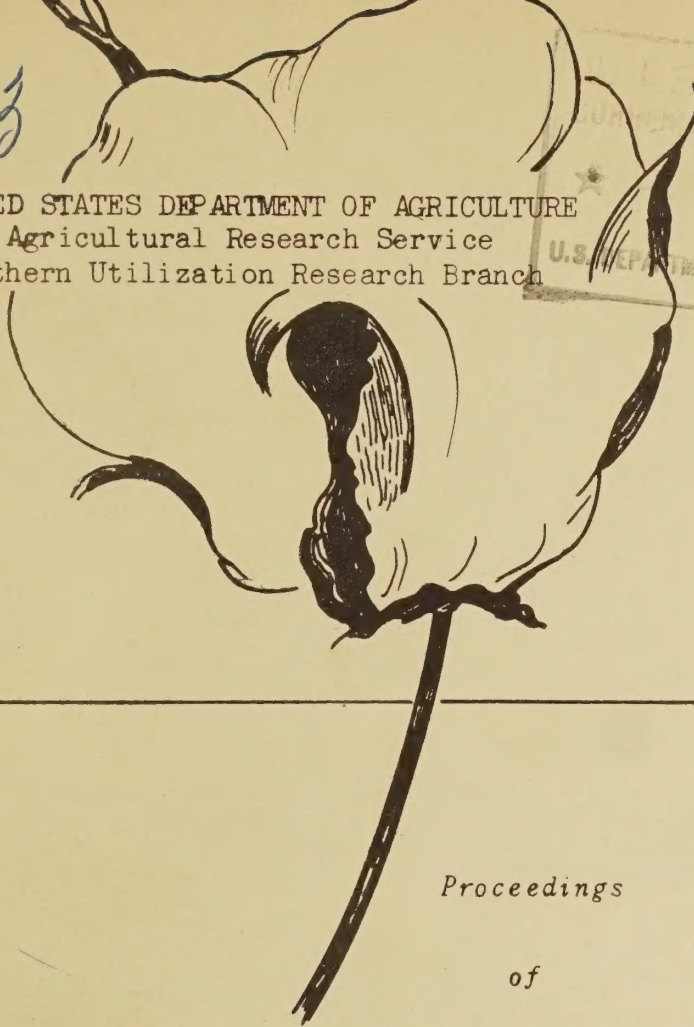
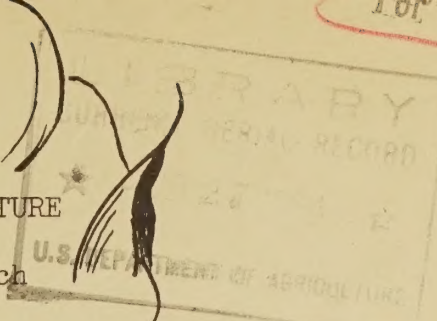


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UNITED STATES DEPARTMENT OF AGRICULTURE
Agricultural Research Service
Southern Utilization Research Branch



Proceedings
of
THIRD CONFERENCE ON PROCESSING AS RELATED
TO NUTRITIVE VALUE OF COTTONSEED MEAL

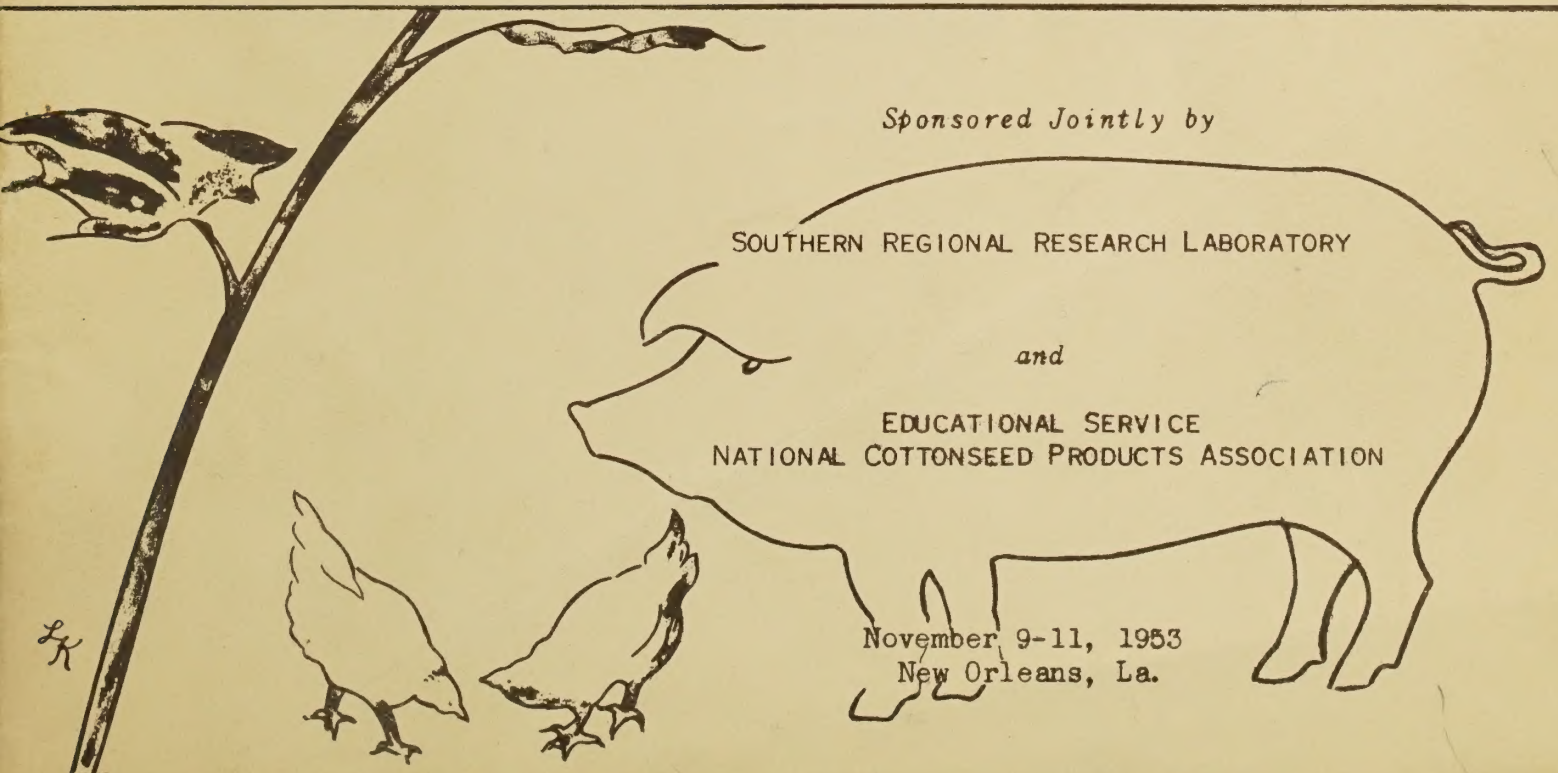
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SOUTHERN REGIONAL RESEARCH LABORATORY

and

EDUCATIONAL SERVICE
NATIONAL COTTONSEED PRODUCTS ASSOCIATION

November 9-11, 1953
New Orleans, La.



FOREWORD

In this report is summarized the information presented at the 3rd conference on processing as related to the nutritional value of cottonseed meal, sponsored jointly by the Educational Service of the National Cottonseed Products Association and the Southern Regional Research Laboratory. The conference was held at the Southern Laboratory in New Orleans, Louisiana, November 9-11, 1953.

The participants met to review the information obtained on the properties of cottonseed meals produced by the newer methods of commercial processing and to evaluate the results of all current nutritional investigations of cottonseed meals. It was desirable to establish the current status of cottonseed meal utilization in swine and poultry feeds so that an effective program for future research might be planned.

This report contains abstracts and discussions of all data reported as well as a summary of the results and future objectives of investigators in this field.

Further details may be obtained by writing to the Southern Regional Research Laboratory, 2100 Robert E. Lee Blvd., or to persons listed on the program.

It would be appreciated if none of these proceedings are reproduced or used without consulting the author or organization concerned as they are preliminary in nature.

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OPENING REMARKS

C. H. Fisher, Director
Southern Regional Research Laboratory
New Orleans, La.

Again it is my privilege and pleasure to welcome you to a conference on processing as related to the nutritional value of cottonseed meal. I express the feelings of the entire staff of the Southern Laboratory when I say that we are delighted to have you with us and that we hope that your visit will prove to be profitable and completely satisfactory. We appreciate your willingness to assemble here to exchange information on cottonseed processing. We are certain that this Conference, like its predecessors, will be of great benefit to research on cottonseed utilization.

It has been said that scientists in the United States have made at least two great contributions to the philosophy and methodology of research. These contributions were achieved by demonstrating that fundamental information can be put to work by skillful applied research and that complex problems frequently can be solved best by cooperative research. Of the examples that might be cited to illustrate the value of cooperative work, the program on cottonseed research to be discussed at this conference is one of the best. As you know, many individuals and organizations are working together in a well coordinated effort to improve the processing and utilization of cottonseed. We in the Southern Laboratory are happy to be a member of the splendid team doing this work, and we wish to express our gratitude for the excellent contributions being made by State Experiment Stations, universities, industrial and commodity organizations and other Federal agencies. We wish particularly to thank Mr. A. L. Ward and other members of the National Cottonseed Products Association for their friendly and effective cooperation.

In conclusion, I should like to remind you that the Southern Laboratory conducts research on several southern crops and that you are cordially invited to visit us at any time to discuss research and offer suggestions.

A. L. Ward, Director
Educational Service, National Cottonseed Products Association
Dallas, Texas

I am grateful to have the pleasant privilege to join Dr. C. H. Fisher and the Southern Regional Research Laboratory staff in welcoming you and expressing to each of you the appreciation of the cottonseed crushing industry for your attendance and participation in this Third Cottonseed Meal Nutrition Research Conference.

We are brought together here today by a common objective - - - that of efficient and unrestricted use of cottonseed meal in the rations of poultry and swine. As you so well know, many years of outstanding performance have won for this protein concentrate a highly regarded position in the ration of dairy and beef cattle and sheep. That favored position, established by research and experience, caused Dr. Rufus Cox, an outstanding livestock specialist of the Mid-West to say, "It has been one of the true pioneers which has helped to develop the range country of the United States. It has been a vital link in the nutrition of range cattle and sheep".

The fact that cottonseed meal has not been able to offer the same unrestricted usage to poultry and swine is a challenge to your research initiative. Each of you deserves congratulations and the appreciation of the cotton grower, seed processor, feed manufacturer and feeder alike because you have accepted the challenge to uncover the true value of the protein of cottonseed meal for poultry and swine. Because of your individual and cooperative contributions to this task, we are assured that the crop production and feeding economy of the South, Southwest and Far West will be enriched.

During the 1952-53 feeding year, approximately 65% of all supplemental protein was supplied by oilseed meals. This fact, alone, justifies our constructive program to improve the value of one of the most important oilseed meals and widen its usefulness.

During that feeding year, the combined dairy, beef cattle and sheep consumption of supplemental protein equaled about six million tons of protein concentrate on an oil meal equivalent basis. Naturally, cottonseed meal supplied much of this protein. However, because of reasons which this research program seeks to overcome, cottonseed meal supplied very little of the approximately seven and three-quarters million tons required by poultry and swine.

You, as a research worker, know that the poultry and swine feeder, in the cotton-growing and surrounding states, is not content to be denied unrestricted use of cottonseed meal. Such use would give to these producers the equal opportunity and even an additional advantage over producers in other areas.

Just how important this research program is to the poultry and swine feeder is obvious when we compare prices of cottonseed meal and soybean meal, the vegetable protein which is most commonly used in the rations fed simple-stomached animals. The October 24 edition of "Feedstuffs" magazine quoted prices for the two which indicated that soybean meal brought \$17.00 per ton more than cottonseed meal in Los Angeles; \$16.00 more in Fort Worth; and \$15.00 per ton more in Memphis. Feed manufacturers and feeders recognize that the freight charges which build up the cost of soybean meal in the cottonseed meal country demand our best efforts to produce a cottonseed meal which will remove this disadvantage.

The research progress you will report here during this Conference and the plans you will make, help to insure the further development and stability of the agricultural and processing economy of the Cotton Belt and earns for you the gratitude of all those who seek progress.

HISTORY OF COOPERATIVE RESEARCH PROGRAM AND REVIEW OF PREVIOUS CONFERENCES

A. M. Altschul
Southern Regional Research Laboratory
New Orleans, La.

It is the purpose of this presentation to set the stage for this conference by reviewing the pertinent events occurring within the last five years. I might start with a brief chronology.

As early as 1941, considerable effort involving research on the pigment glands and pigments of cottonseed was expended at the Southern Regional Research Laboratory by Dr. Charlotte Boatner Miller and her associates. The results of this work indicated a need for a re-evaluation of the effect of processing upon the quality of cottonseed meal, which could come only through a concerted effort involving chemists, commercial processors, the cottonseed industry, and nutrition experts. In December 1948 at a meeting of the New Food and Feed Committee of the Department of Agriculture, plans were made for cooperative research between the Southern Regional Research Laboratory and nutrition experts in the Bureaus of Animal Industry, Dairy Industry, and Human Nutrition and Home Economics to determine the nutritive value of cottonseed meals produced under specified conditions of processing. These experimental meals were prepared in a cottonseed oil mill in South Texas which had been cooperating very closely with this Laboratory in previous processing experiments.

At about the same time, the Southern Regional Research Laboratory and the Educational Service of the National Cottonseed Products Association initiated an informal cooperative program which has continued and prospered. As people of various talents become involved in this cooperative work, it became clear that periodic meetings to serve as a clearing house of information and to focus attention on the unsolved problem would be a great asset to the progress of the work. For this reason, formal conferences were held in November, 1950, and in November, 1951, and several informal conferences were held with smaller groups during the intervening period. This third conference is a result of the decision made by an informal committee with Dr. C. M. Lyman as chairman to review recent research and to decide on a future research program. The participants in this cooperative program, including the National Cottonseed Products Association, its educational service, and fifteen of its affiliated mills, State, Federal and Industrial Nutritional Laboratories, and the Southern Regional Research Laboratory, are not bound by formal organizations but rather by an intense mutual interest and a sincere desire to achieve the maximum usefulness for cottonseed meal.

I do not wish to imply that all or even most of the productive work on cottonseed meal was done in this cooperative program. This would be doing a great injustice to the excellent work of pioneers in the fields of cottonseed meal nutrition and gossypol chemistry and of individual investigators presently working outside of this cooperative program. All we may say is that the work involved in this cooperative program has been significant as attested by your presence here and by the very representative groups present at the first two conferences. It is our hope that this community of interest which has made this extensive cooperation possible will continue until the major problems have been solved and the cottonseed industry is well on its way to more extended utilization of higher quality products.

I would like to direct your attention to the history of ideas as they developed through the cooperative efforts of all participants in this program. In the past, cottonseed meal has been used primarily in beef and dairy cattle feeds but restricted in its use in swine and poultry feeds. The problems, therefore, were to develop chemical methods of identifying those meals of high utility, and, at the same time, to maintain the high quality of the oil or, if possible, to improve it.

Having established the problem, two questions arose:

1. Can the problem be solved, and
2. How can it be solved?

The history of this cooperative program is really a history of attempts to answer these questions or to reformulate them in such a way that they can be investigated experimentally.

In trying to answer these questions there have been four major cooperative efforts in which we at the Southern Regional Research Laboratory have been privileged to participate. An understanding of these efforts is necessary to serve as a basis for the discussions of the next few days.

1. ESTABLISHMENT OF VARIABILITY OF COTTONSEED MEALS.

Cottonseed meals have been considered to be standard materials of uniform nutritional properties, regardless of point of origin or conditions of processing. If this were indeed true, there would be very little hope of improving the nutritive value and utilization of cottonseed meal. It was necessary, therefore, to determine whether cottonseed meals were uniform or variable depending upon conditions of processing. The Southern Regional Research Laboratory in cooperation with several commercial cottonseed mills prepared samples of meals by varying conditions of processing, such as, temperature of cooking, the heat input in the press, the conditions of oil extraction, etc. These meals were distributed widely for nutritional investigation and comparison with a standard cottonseed meal. The standard meal was produced in the pilot-plant of the Southern Regional Research Laboratory by successive extractions with hexane and butanone. This meal had low free and total gossypol contents and was subjected to a minimum amount of heat in evaporation of the solvent.

The results of nutritional investigation on these samples showed clearly that the variability in quality of cottonseed meal was a function of processing conditions. Furthermore it has been established that some of the cottonseed meals have rather high nutritive values and can be fed in unrestricted quantities to swine and growing poultry. It is apparent from these results that the problem of improving the nutritive value of cottonseed meal can be solved.

2. DETERMINATION OF THE ROLE OF FREE GOSSYPOL.

There are two factors determining the utilization of cottonseed meal for swine and poultry: (1) the presence of materials that interfere with growth and (2) the damage to the protein during processing. It was necessary to separate these two factors and to determine the contributions of each to the utility of cottonseed meal. For this purpose, special rations were prepared containing meals of different levels of free gossypol and others to which pure gossypol had been added.

The Southern Laboratory distributed quantities of gossypol for determination of its toxicity and its effect when added to feeds. Mixtures of cottonseed meals, containing different quantities of free gossypol, prepared at this laboratory by blending various meals, were also furnished to nutrition investigators.

There are some areas of general agreement now on the levels of free gossypol that affect the growth of swine and poultry. It would seem that cottonseed meal containing 0.03 and 0.04% free gossypol can be fed safely to swine and growing poultry if they constitute 30% of the diet. Meals containing more free gossypol are suspect although the critical level is not clearly defined.

A very important problem for consideration is the effect of gossypol on egg production and egg quality. A considerable amount of work in this field has been done using mixed meals and adding pure gossypol. Some of this work has been reported previously, and we have scheduled further reports at this meeting.

While we believe that the general picture is much clearer now, as a result of cooperative work on swine and poultry, there is room for continued research on the role of free gossypol in the nutritive value of cottonseed meal.

3. EVALUATION OF COMMERCIALY AVAILABLE AND EXPERIMENTAL MEALS.

Having established tentatively the fact that levels of gossypol below 0.04% do not interfere with growth, it is possible then to examine independently the effect of processing conditions on the quality of the meal. This was done for quite a variety of commercial meals as well as modified meals produced in commercial plants. All of these meals had low free gossypol content and therefore the variation in nutritive value of these meals could be largely attributed to changes in protein value. The commercial

screw-press meals, although low in free gossypol, have suffered considerable protein damage due to heat. In order to overcome this difficulty, special screw-press meals were prepared at lower temperatures or under modified cooking conditions similar to those used in the hydraulic press method. The commercial as well as modified meals are the Series 5, 12, 14, and 15 meals which have been distributed to many nutritionists and reported in a number of publications. Nutritional results indicated that the screw-press meals cooked at temperatures below 200°F. prior to pressing are generally nutritionally superior and approach the standard meal described above. Attempts to modify the commercial screw-press operation to produce low temperature-cooked meals have thus far not been very successful.

Very recently we have made a survey of commercial meals produced by the pre-press, solvent-extraction process. We have available all of the chemical and processing data and some of the nutritional data, all of which will be presented as part of the program. High-speed and high-capacity screw-presses have recently been developed. It would be interesting to obtain similar information on the quality of the product produced from such presses as well as from other new commercial operations.

The Southern Laboratory, in cooperation with the cottonseed industry, is continuing to distribute meals to nutrition investigators. The latter should insist on having processing information on all the cottonseed meals they test and should become thoroughly familiar with those meals which seem to be approaching the desired nutritional properties. Then they can advance with confidence to research on the mixing of cottonseed meals with other meals, on the effect of supplements (amino acids, etc.), bound gossypol, and other constituents of the diet. They can better approach incidental problems such as dermatitis in swine if they have at hand a clear definition of the cottonseed meal used.

4. FUNDAMENTAL RESEARCH ON THE COOKING OF COTTONSEED MEATS.

The efforts to produce improved meals by minor modification in the present operation have fostered the belief that the commercial production of meals of higher quality depends upon a greater understanding of the chemistry of the cooking process. For this reason the Southern Regional Research Laboratory initiated laboratory research on the fundamentals of the cooking process about a year ago. This research replaced former studies in cooperation with cottonseed oil mills on modern modification of processing conditions. You will hear a complete report of the progress of our research on cooking at this meeting. I might say that we have reason, as a result of the progress to date, to be optimistic that not only good meal but also high quality oil can be made by modification in cooking conditions. A corollary of this work is our interest in rapid biological tests for nutritive value and in developing a chemical measure of heat damage that will parallel the changes in nutritive value.

I should like to close these comments by listing some of the questions, the answers to which might be reasonably expected to emerge from this meeting.

We should like to know whether commercial meals are now being produced which are so superior to those generally produced that they can be considered for extended utilization in animal feed. For example, how do the pre-press solvent-extracted meals compare to other meals? Is there a way of identifying these meals so that the consumers can specify the type of meal they want for this extended utilization?

What are the chances of the entire industry being able to produce superior meal? The answer to this may lie in the result from fundamental research on cooking and possible commercial application. Also of interest is the chemical approach by the Buckeye Cotton Oil Co. which no doubt will be mentioned during the course of the discussion.

The success of the program depends largely on the development of a rapid measure of nutritive value; research on chemical estimation has been supported by a fellowship of the National Cottonseed Products Association. Are there any rapid biological methods for measuring nutritive value?

In most of our discussions, cottonseed meal has been considered as the sole source of protein in animal rations, but if it were used in combination with other proteins, could its utilization in swine and poultry diets be expended in the immediate future? How significant is the limitation due to low concentrations of amino acids?

Egg-yolk discoloration remains one of the most difficult problems to solve. What is the minimum free gossypol content necessary to eliminate egg-yolk discoloration? What approach can we make to the solution of this problem?

No doubt many other questions will occur to you during the course of this meeting. It is our hope that on the last day of this conference, we will be able to evaluate each of the subjects discussed and list our results and objectives in order to serve as a guide for those who wish to continue this type of work, either as a part of a group effort or individually.

Seminar I - USE OF COTTONSEED MEAL IN PRACTICAL CHICK AND BROILER RATIONS

Chairman - J. A. Kime, New Orleans, La.

USE OF COTTONSEED MEALS IN PRACTICAL CHICK AND BROILER RATIONS

J. R. Couch

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Sherwood and Couch (1950) reported that cottonseed meals with low gossypol content could be used as the protein supplement in broiler rations when supplemented with 1 percent dl-lysine.

Milligan et al. (1951) conducted a series of experiments and concluded that when cottonseed meal was used as the only protein supplement in a chick diet that a total of 1 percent l-lysine was required during the first six weeks for best growth. It was necessary to add 0.8 percent dl-lysine monohydrochloride to the diet in order to meet the lysine requirement when cottonseed meal served as the only protein supplement.

Milligan and Bird (1951) reported that improvements in cottonseed meal processing might make it possible to manufacture a cottonseed meal that was equal to soybean oil meal in feeding value for growing chicks.

Machlin, Denton and Bird (1952) concluded that lysine was the only amino acid that was deficient in a diet composed largely of corn and cottonseed meal when fed growing chicks up to six weeks of age.

Reed, Quisenberry and Couch (1953) conducted an experiment in which one-half of the protein supplement was composed of soybean oil meal and the remaining half was supplied by an experimental cottonseed meal (Series 12) produced under the supervision of the Southern Regional Research Laboratory. In this instance, the growth of the birds was increased significantly and the feed efficiency was improved when the diet was supplemented with 0.05 percent dl-methionine. The increase in growth and improvement in feed efficiency obtained with the cottonseed meal-soybean oil meal diet was almost identical with that which resulted from the feeding of soybean oil meal as the principal protein supplement.

Couch (1950) reported that a group of cottonseed meals (Series 5) prepared by the Southern Regional Research Laboratory proved to be unsatisfactory when such meals were used as the only protein supplement in an all-vegetable protein diet for broilers.

Bird (1951) reported that experimental screw-press cottonseed meals and a commercial sample of cottonseed meal replaced soybean oil meal satisfactorily in a commercial type, high efficiency broiler ration. Growth reported by Bird was entirely satisfactory. Unfortunately, the diet used in this study was not given. It is presumed that cottonseed meal was not used as the only protein supplement, but was probably added in combination with soybean oil meal, fish meal and other protein concentrates, since the ration was designated as a commercial type, high efficiency broiler ration.

German, Reed and Couch (1951) summarized the data from the Texas Station at the Second Conference on Cottonseed Processing where selected experimental meals from Series 5 through 14 had been used in broiler feeding tests. It was found that cottonseed meal could be used to replace one-half of the soybean oil meal in an all-vegetable protein ration. Growth obtained through the use of the one-half cottonseed meal and one-half soybean meal ration was significantly greater than that obtained when soybean oil meal was used as the only protein concentrate. It appeared from this data that this mixture of protein sources might be superior to soybean oil meal alone for use in broiler feeds. The increase in growth which resulted from replacing one-half of the soybean oil meal with cottonseed meal quite possibly could have been due to the amino acid composition of the cottonseed meal or to the fact that the cottonseed meal might possibly contain an unidentified growth factor. Growth was further enhanced in these studies through the addition of fish meal (3 percent) to the rations containing soybean oil meal alone or to those containing a mixture of soybean oil meal and cottonseed meal.

Data obtained during the past two years from a number of laboratories have shown rather consistently that there is an unidentified growth factor in fish meal, liver meal and fish solubles. Such factor has been designated by some as the "fish factor".

Lyman, Chang and Couch (1953) reported on a series of extensive experiments conducted in the laboratory of the Department of Biochemistry and Nutrition of the Texas Agricultural Experiment Station in which it was shown that the total gossypol content of the cottonseed meals tested appeared to be the most important factor in determining the nutritional value of the protein.

In summary, it might be stated that all research data to date have shown that cottonseed meal is deficient in lysine. This amino acid is a limiting one in the poultry feeding field since grains, wheat by-products, and other ingredients low in protein used to make up the primary portion of poultry feeds are also low in this amino acid. Soybean oil meal is high in lysine. There is little question but that a mixture of soybean oil meal and cottonseed meal could be selected for use in poultry rations.

It is an established fact that gossypol combines with lysine to form a complex that is not acted upon by the enzymes in the digestive tract of the chick; thus, the gossypol-lysine complex is excreted and the amino acid is lost to the bird. Gossypol is not only toxic when fed in high concentrations, but is also the most important limiting factor in tying up the critical amino acid, lysine.

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USE OF COTTONSEED MEALS IN PRACTICAL CHICK AND BROILER RATIONS

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Cottonseed meals produced by various processes have been used in chick rations as protein supplements, alone and in combination with soybean meal. Each 100 pounds of the basal rations used in the various tests contained the following: fish meal, 2 lbs.; dried whey, 2 lbs.; salt, 0.5 lbs.; steamed bone meal, 2 lbs.; and limestone, 1.5 lbs. plus added vitamins, an antibiotic, methionine and a coccidiostat. Soybean meal was used as the protein supplement in the control ration. In these tests degossypolized solvent, screw-press, pre-press solvent and hydraulic cottonseed meals were used.

Of the various cottonseed meals tested, results have been most favorable with the degossypolized solvent extracted meal. In one test, this type meal when used alone in the diet was equal (statistically significant) to the soybean meal control ration. When used in combination with soybean meal (1:1), all types of cottonseed meal, except the hydraulic meals in certain instances, have given satisfactory growth results. The pre-press solvent extracted cottonseed meal, although not used in direct comparison with the screw-press meal, appears to be of higher nutritional value than the screw-press type. The hydraulic cottonseed meal is of inferior nutritive value as compared with cottonseed meals processed by other methods.

Summary of results on New Hampshires 10 weeks of age - (2-19-53 to 4-15-53)- 20 birds per duplicate lot.*

Supplement	Average weights and feed per pound gain			
	Pen A		Pen B	
	Feed per		Feed per	
	Weight	lb. of gain	Weight	lb. of gain
	gms	lbs	gms	lbs
Soybean Meal	1437	2.50	1404	2.51
Screw-press CSM	1207**	2.84	1306**	2.80
Soybean Meal plus Screw-press CSM	1463	2.54	1373	2.68
Degossypolized solvent CSM	1389	2.70	1366	2.83
Soybean Meal plus Degossypolized solvent CSM	1426	2.65	1509	2.62
Hydraulic CSM	1258**	2.89	1090**	2.90
Soybean meal plus Hydraulic CSM	1448	2.50	1504	2.51

*Reported - Poultry Science, August, 1953

**Significant at the 0.01 level.

Analysis of above meals.

Product	Protein %	Fat %	Gossypol*	
			Free %	Total %
Soybean Meal	45.56	0.97	-	-
Degossypolized solvent CSM	41.06	1.64	0.041	0.38
Screw-press CSM	42.00	3.58	0.029	0.70
Hydraulic CSM	41.63	8.32	0.064	0.90

*Gossypol determinations made by Southern Regional Research Laboratory.

Results of feeding various cottonseed meals in chick rations at 18% and 21% protein levels to White Plymouth Rock broilers for eight weeks (4-21-53 to 6-16-53) 20 birds per duplicate lot.*

Supplements	Average weights and feed per pound of gain			
	Pen A		Pen B	
	Weight gms	Feed per lb. of gain lbs	Weight gms	Feed per lb. of gain lbs
<u>21% protein rations</u>				
Soybean Meal	909.7	2.51	909.2	2.35
Hydraulic CSM	416.3	2.82	413.1	2.90
Degossypolized solvent CSM	832.4	2.48	759.7	2.43
Pre-press solvent CSM	800.2	2.55	784.6	2.50
<u>18% protein rations</u>				
Soybean Meal	853.9	2.24	901.2	2.30
Hydraulic CSM	424.2	3.28	388.9	3.30
Degossypolized solvent CSM	793.8	2.37	759.3	2.43
Pre-press solvent CSM	687.6	2.66	681.4	2.62

*Unpublished data - confidential

Analysis of cottonseed meals.

Product	Crude Protein	Fat	Gossypol	
	%	%	Free %	Total %
Degossypolized solvent CSM	44.56	2.27	0.41*	0.63
Hydraulic CSM	44.81	7.04	0.21	1.04
Pre-press solvent CSM	43.31	0.82	0.024	0.80

*Redetermination showed this value to be 0.05%.

Results of feeding various cottonseed meals in combination with soybean meal in chick rations at 21% protein levels to New Hampshire broilers for eight weeks (5-5-53 - 6-30-53) 85 chicks per lot.*

Supplement	Average weight and feed per pound of gain	
	Weight gms	Feed per lb. of gain lbs
Soybean Meal	763.9	2.75
1/2 Soybean 1/2 Hydraulic CSM	562.5	3.19
1/2 Soybean 1/2 Degossypolized solvent CSM	711.4	2.83
1/2 Soybean 1/2 Pre-press solvent CSM	706.2	2.86

*Unpublished data - confidential.

DISCUSSION

Discussion Leader - W. C. Sherman, Ralston Purina Company, St. Louis, Mo.

- Craig: In connection with the data reported by Dr. Morgan, I would like to point out that the value of 0.41% free gossypol* in the degossypolized meal from Buckeye Cotton Oil Company must be erroneous as the free gossypol content of all degossypolized meals produced by Buckeye is carefully controlled at a level of less than 0.04%. The results of the feeding tests reported by Dr. Morgan showed that this meal was one of good nutritive value; if the free gossypol content were actually 0.41%, the chicks would probably have died.
- Hoffpauir: This analysis for free gossypol was made at the Southern Regional Research Laboratory by the method of Pons and Guthrie. We realized that these values for the degossypolized meals were far too high, but it appears that the meal which was degossypolized by the method of processing used at the Buckeye Cotton Oil Company contained a material which enhances the color of the gossypol-p-anisidine reaction product measured by this method of analysis. It is possible that any gossypol derivative remaining in the meal is hydrolyzed under the conditions of this method and selectively reacts with p-anisidine.
- Sherman: The talks which have just been presented raised certain questions about lysine which I feel we should discuss: (1) Must lysine be added to cottonseed meal? (2) Can the lysine in soybean meal make up the deficiency of lysine in cottonseed meal if the two are mixed in equal proportions in the diet? (3) Is the lysine in cottonseed meal more readily available than that in soybean meal?
- Lyman: Some cottonseed meal samples which we have tested for lysine availability have equaled soybean meal but even in such cases, added lysine increased the nutritive value. Added lysine had a greater effect on nutritive value in the case of poor meals.
- Sherman: Is the average value for lysine in cottonseed meal greater than half that for soybean meal?
- Lyman: Yes, in fact, recent analyses of the two have shown that soybean meal contains only a small amount more lysine than cottonseed meal.
- Couch: Cottonseed flour contains 4.64% lysine, whereas soybean flour contains 5.83%.
- Rusoff: Do the fish meals used by Dr. Morgan in the basal diet contribute lysine?
- Morgan: Yes.
- Colby: From the point of view of the chemical manufacturer, certain questions must be answered concerning lysine addition to mixed feeds before production of lysine for this purpose can be considered: (1) Is supplemental lysine really needed? (2) If so, what is the minimum level required? (3) If there is an increase in nutritive value of feed effected by the addition of lysine, what can the feeder afford to pay for the lysine? (4) Can lysine be commercially produced at a price set by the feed industry?
- Sherman: Would the synthetic lysine be dl-lysine?
- Colby: For this purpose it would probably be l-lysine as the d-isomer is not utilized nutritionally.
- Watts: In order to compare nutritional results, there must be some common basis with regard to intake of feed. Morgan's data show differences in consumption for the different rations tested. Could these differences be due to the palatability of the ration rather than to the protein quality?

*Redetermination showed the free gossypol content to be 0.05%.

- Sherman: The question of palatability has been discussed considerably in other groups and has led to many new investigations to evaluate its importance in nutrition.
- Fincher: Does the chicken select food on the basis of palatability or because of its appearance?
- Couch: Chickens like the appearance of yellow corn, which does not improve growth.
- Sherman: The consistency of the meal is important. Chicks will not eat a finely powdered diet.
- Couch: Although the particle size and consistency of feed can be regulated so that the chick cannot select appealing portions, equipment for preparing homogeneous feed is not available to many educational institutions. If it were, these nutritional results would be of more significant value.
- Morgan: Could the differences in the consumption of feed containing cottonseed meal be due to some physiological effect rather than palatability?
- Eagle: Thus far, the question of toxicity has been omitted from the discussion. The physiological action of gossypol in the rat involves paralysis of the pyloric sphincter which causes the stomach to remain full and reduces consumption of the feed to a minimum causing the animal to starve to death. The free gossypol level in cottonseed meal, therefore, cannot be used as a criterion for toxicity to rats.

Seminar II - DISCUSSION OF PIGMENTS AND AMINO ACIDS DISTRIBUTION IN COTTONSEED

Chairman - H. D. Fincher, Anderson-Clayton and Co., Houston, Texas

PIGMENT DISTRIBUTION IN COTTONSEED PRODUCTS AND A DEFINITION OF TERMS USED

A. M. Altschul

Southern Regional Research Laboratory
New Orleans, Louisiana

The cottonseed kernel contains 1 - 2% pigment glands by weight. About 50% of the weight of the glands is made up by pigments. Ninety percent of this pigment fraction is gossypol. Gossypol is a yellow, physiologically active pigment. Dr. Eagle and other workers have shown that the physiologically active factor which is deleterious is found in the pigment glands. They have shown that pure gossypol is toxic. We know that 100% of the gossypol in the kernel can be extracted by aqueous acetone.

The cottonseed kernel can be treated in many ways. From the investigations of the samples furnished for swine and poultry feeding studies, we have followed the distribution of gossypol in the processed cottonseed products. Some of the gossypol is inactivated and remains in the meal. A portion of the original content is destroyed and cannot be accounted for by the present analytical methods of determining total gossypol. Some of the gossypol goes into the oil, the amount depending upon the method of processing. For example, in pre-pressed oils, the gossypol content is about 0.7%, whereas hydraulic pressed oil may contain 0.2%.

By free gossypol we mean the gossypol extracted by 70% aqueous acetone during a one hour extraction period. A hexane-extracted cottonseed meal has a free-gossypol content of about 0.6%. This high free-gossypol content can be explained by the fact that the pigment glands are not ruptured by hexane, and the gossypol is unreacted. Of course, some of the gossypol will go into the solvent-extracted oil. Total gossypol is that amount determined by a hydrolysis method that requires heat treatment and oxalic-acid. This hydrolysis frees the reacted gossypol of the meal. Bound gossypol is the difference between the contents of free and total gossypol.

AVAILABILITY OF AMINO ACIDS IN COTTONSEED MEALS

C. M. Lyman

Department of Biochemistry and Nutrition,
Texas Agricultural Station, College Station, Texas

Protein material consists of 22 different building blocks, or so-called amino acids. Eight or ten of these amino acids cannot be manufactured from assimilated protein by the animal; they are called the essential amino acids. In the discussion of cottonseed meal, we are concerned with three of these: lysine, tryptophane, and methionine. The usual animal rations contain the other essential amino acids in sufficient amounts.

If the amino acids are not in a form available for use, they are not of value to the animal. Lysine is the limiting protein building block in cottonseed meal. A relative measure of the availability of lysine can be obtained from feeding studies with rats by the determination of the relative lysine contents of the ration and of the feces. The difference between these values is the amount of available lysine. In cottonseed meal, only 60% of the lysine is available. Gossypol is related to the unavailability of lysine in the meal. It appears that the inactivation of gossypol parallels the decrease in availability of lysine. In the data reported at this conference, we must keep in mind the two separate functions of gossypol, its toxicity and its binding capacity. The animal can only use the natural form of lysine, or L-lysine. Cottonseed meal contains almost as much lysine as soybean meal, but not all of it is available.

DISCUSSION

- Bryson: Does thiamine influence the nutritional evaluation of cottonseed meals?
- Altschul: The thiamine contents of the meals distributed for feeding studies have been determined. It was found that thiamine was destroyed by heat during processing, but the changes in thiamine contents do not seem to be directly associated with the changes in nutritive value.
- Bryson: Is thiamine a vitamin or an amino acid?
- Lyman: It is one of the vitamins which are present in traces in the meal. The stability during processing of methionine and arginine, two of the amino acids present in the meal, is of more interest to nutritionists.
- Bryson: Would lysine bind to free gossypol if added before processing of the seed or at the time of feeding?
- Lyman: Lysine, when added at the time of feeding, does not change the gossypol toxicity. Added lysine apparently does not react with free gossypol but either remains unchanged or is tied up by the sugar present in the diet. Gossypol evidently is selective in its reaction with the native lysine of the protein.
- Bryson: Does the lysine content or the protein quality differ in prime cottonseed and in off-grade seed?
- Lyman: In all types of seed, the percent lysine in the protein is the same, although the nitrogen content may vary. The development of free fatty acids in off-grade cottonseed does not change the lysine content or the protein quality.
- Sandahl: Is there an ideal proportionality of amino acids for all animals?
- Lyman: The necessary amino acid distribution pattern is the same for all non-ruminants.
- Altschul: If it were possible to get the amino acids of the protein into the pattern ideal for the animal, our problem would be solved.

Seminar III - STATUS OF COTTONSEED MEAL FOR SWINE

Chairman - H. D. Fincher, Anderson-Clayton and Co., Houston, Texas

COTTONSEED MEAL FOR SWINE

N. R. Ellis

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Beltsville, Maryland

In reviewing progress of research on the feeding of cottonseed meal to swine, several phases need to be considered; namely, gossypol toxicity, use in the gestation-lactation diet, and use in the growing ration.

A tabulation of published data on the levels of free gossypol either tolerated by pigs or at which ill effects occur shows a considerable range of values. Some workers have observed ill effects on diets containing as little as 0.01 percent, whereas others have seen no toxic effects up to 0.024 percent (U.S.D.A.). On the average, a level of 0.02 is suggestive of the possible threshold point, since this amount has been used with safety by a number of investigators. In the feeding experiments of recent years, utilizing cottonseed meals of low gossypol content, toxicity effects have been a very minor problem. It can be concluded, therefore, that meals containing 0.03 to 0.05 percent free gossypol can be used with safety up to 20 percent of the diet or even somewhat higher. Obviously, more experiments are needed to fix the exact limit of free gossypol consumption and the other dietary factors surrounding safe usage. For the present, perhaps 0.01 percent is the maximum that can be used in a diet with safety.

Several reports have appeared on the use of cottonseed meal as the main protein concentrate in diets of sows during gestation and lactation. While reasonably good litter performance has been obtained there is room for improvement, especially in weaning weights of pigs as well as general thriftiness.

A large number of experiments have been conducted with growing pigs. Most of the southern experiment stations along with the U. S. Department of Agriculture have participated in recent years in testing meals for protein quality many of which were supplied through the Southern Regional Research Laboratory. Feeding results have ranged from fair to very satisfactory. Various nutritional problems have been encountered. Experience gained from several feeding trials at the Agricultural Research Center will be used to illustrate the problems encountered and the progress made.

In the initial trial at Beltsville, Maryland, a screw press meal performed about as well as a check lot of soybean meal and better than a sample of hydraulic meal. The next two tests attempted the evaluation of the effects of different cooking temperatures (Series 5 meals). While the meals prepared at lower temperatures appeared to be better than those at higher temperatures, none quite measured up to the results from the check lot on soybean meal. Complicating the results were numerous cases of lameness in the pigs fed cottonseed meal in contrast to general freedom from this disorder on soybean meal. Pantothenic acid deficiency, first suspected as responsible for lameness, was eliminated as the responsible factor. A further test established the point that the pigs were suffering from rickets. In appraising the calcium, phosphorus and vitamin D relationships, the phytin in the cottonseed meal was naturally suspected as playing a role.

A factorially designed test to evaluate the rates of calcium, phosphorus and vitamin D levels next brought into focus a third effect of diet, i.e., a dermatitis, in addition to rate of gain and lameness. When total calcium and phosphorus were held near to the recommended levels of the National Research Council, severe lameness was encountered; the gains were rather poor, but no dermatitis was observed. An increase in phosphorus alone gave a better growth response but did not eliminate lameness and allowed some dermatitis to appear. Addition of both calcium and phosphorus eliminated lameness but apparently aggravated the dermatitis as well as retarded growth. In the corresponding four groups given additional vitamin D, lameness was eliminated only in the group given extra phosphorus or both calcium and phosphorus, whereas dermatitis was eliminated in the other two groups (no extra calcium and phosphorus or

extra calcium only). Gains were good in two lots; namely, added vitamin D only and added phosphorus and vitamin D.

The role of wheat as a partial replacement of the corn in the diet to supply phytase was next tested in the presence of high versus low calcium and phosphorus and of high versus low vitamin D. The results again showed the conflict in rate of gain, lameness and dermatitis. At the same time the wheat contributed very little to correction of the troubles. While skin parasites have not been ruled out as factors in dermatitis, the pattern of incidence appears to be associated with nutrition. Because a diet, producing at the same time a high growth rate and freedom from dermatitis and lameness, has not yet been achieved, the evaluation of protein quality of cottonseed meals thus remains compromised.

Finally, tests on phytin involvement have rather definitely established the point that feeding this substance to pigs resulted in faulty mineral metabolism and consequent lameness. When the dietary phosphorus other than the phytic acid phosphorus is inadequate, rickets results in presence of a moderately low level of vitamin D.

While the swine nutritionists at the other experiment stations have not been confronted with so complex a problem as experienced at Beltsville, there is much concern over the dermatitis question. Probably, the vitamin D level has been compensated for by greater exposure of pigs to sunshine. The whole problem of association of rate of growth, rickets and dermatitis in feeding of cottonseed meals needs to be resolved just as rapidly as possible.

THE EFFECT OF COBALT AND ZINC IN PREVENTING DERMATITIS IN PIGS RECEIVING CORN-COTTONSEED MEAL RATIONS

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The Buckeye Cotton Oil Company
Cincinnati, Ohio

Presented by H. L. Craig

The Buckeye Cotton Oil Company
Cincinnati, Ohio

Occasional pigs in dry lot have developed dermatitis when fed rations containing corn and cottonseed meal. The syndrome has also been reported in pigs fed corn-soybean oil meal and corn-peanut meal rations. It has been suggested that the addition of alfalfa meal, meat and fish products, or antibiotics would prevent the condition.

In a previous test in the Buckeye Laboratory, two pigs with severe cases of dermatitis made rapid recovery when their ration was supplemented with cobalt and zinc.

A factorial design experiment was conducted to study the effect of the addition of specially prepared cottonseed fatty acids and a combination of cobalt and zinc on the growth and performance of Duroc-Jersey weanling pigs fed to market weight. The basal ration contained corn, degossypolized cottonseed meal, 3% fish meal, steamed bone meal, ground limestone, iodized salt, manganous sulfate, A and D oil, riboflavin, calcium pantothenate, niacin, choline, Vitamin B₁₂ and aureomycin.

The supplements to the basal rations were as follows: none (4 pigs), 1% specially prepared cottonseed fatty acids (8 pigs), and 2% specially prepared fatty acids (4 pigs). One-half of the pigs in each lot received 1.1 ppm cobalt and 4.0 ppm zinc added to the ration. All pigs were self-fed individually on concrete floors. The results of the test are given in the following table:

<u>Supplement</u>	<u>No. of Animals</u>	<u>Ave. initial weight lbs.</u>	<u>Ave. final weight lbs.</u>	<u>Ave. daily gain lbs.</u>	<u>Feed/100 lbs. gain lbs.</u>
Basal	8	38	151	1.35	314
Basal + Co and Zn	8	39	194	1.85	313
Basal	3*	44	193	1.77	317
Basal + 1% CS fatty acids	7*	40	187	1.75	302
Basal + 2% CS fatty acids	3*	39	196	1.86	318

*One pig developed dermatitis--not included in data.

All of the pigs receiving cobalt and zinc had clear skins and good hair coats, and made excellent gains during the entire feeding period. Three of the eight pigs receiving the basal ration without added cobalt and zinc developed moderate cases of dermatitis. The pigs receiving supplemental cobalt and zinc gained 37% faster than the pigs that did not receive supplemental cobalt and zinc. The average daily gain of the eight pigs receiving supplemental cobalt and zinc was 11% greater (1.85 vs. 1.67) than the five pigs that did not develop dermatitis in the unsupplemented lot. This indicates that the lower gains of the group receiving no supplemental cobalt and zinc was not caused entirely by the three pigs with dermatitis. There was no correlation between the development of dermatitis and the addition of cottonseed fatty acids to the rations.

It is evident from the results that the addition of cottonseed fatty acids had no detrimental effect on growth rate or feed utilization.

PRE-PRESSED SOLVENT EXTRACTED COTTONSEED MEAL
AS A SOURCE OF PROTEIN FOR THE WEANLING PIG

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This study was designed to compare the feeding value of pre-pressed solvent cottonseed meal to that of solvent extracted soybean oil meal for weanling pigs fed in dry lot. In view of recent experimental data indicating that 14 percent crude protein was adequate for such animals when fed corn-soybean oil meal rations, it was further desired to determine if such a level of protein was optimum when cottonseed meal replaced the soybean oil meal. In addition, the supplementary value of DSC (distiller's solubles concentrate), a by-product of the production of Monosodium Glutamate from wheat gluten, was determined. This product was obtained from the Huron Milling Company of Chicago. The amino acid analysis indicated possible value as a supplementary source of lysine. A final objective was to determine the value of adding a high level of aureomycin (60 gms/ton) to the corn-cottonseed meal ration. The study consisted of two trials as shown in Table 1. The two experiments were the same except that the cottonseed meal was from different mills. The meal used in Experiment I was obtained from Sherman, Texas (Lab. No. O-3748) and that of the second experiment came from the Dothan Oil Mill Co. at Dothan, Alabama.

Sixty purebred Duroc and Duroc x Hampshire pigs served as the experimental animals. They were carefully allotted to the various experimental groups according to live weight, breed, and litter. The pigs were maintained in concrete pens which were scrubbed and washed down daily. All rations were fed ad libitum in self feeders.

In addition to corn and soybean oil meal or cottonseed oil meal, all rations contained 1 percent limestone, 0.5 percent steamed bonemeal and 0.53 percent of a trace mineralized salt mixture. The trace mineral mixture included iodine, copper, iron, manganese, and cobalt. Aureomycin in the form of Aurofac 2-A was added to all rations at a level of 20 gms. per ton of feed. Ration No. 4 contained three times this amount. Lederle Fortafeed 2-49C was added to all rations at a level of two pounds per ton of feed. The Fortafeed contributed niacin, riboflavin, pantothenic acid and choline.

Results of the experiments, which covered a period of nine weeks, are presented in Table 1. In both experiments, the corn-soybean meal control ration containing 14 percent protein produced better results than any of the rations containing cottonseed meal. Distiller's solubles concentrate (DSC) and a high level of aureomycin were ineffective supplements. In the first experiment the higher level of protein (Lot 5 vs. Lot 2) appeared to be less satisfactory than the lower protein ration. In the second experiment, however, the high protein cottonseed meal ration promoted decidedly better growth than did the low protein rations. The complicating effect of dermatitis in Experiment I may explain this difference.

Dermatitis

Studies during the past four years have indicated a rather high incidence of dermatitis among young pigs fed corn-cottonseed meals in dry lot. The syndrome is not peculiar to corn-cottonseed meal rations alone, since it has been observed many times on corn-peanut meal and corn-sunflower-seed meal rations, and in a very few instances on corn-soybean oil meal rations. The disturbance is characterized by thick scabby incrustations which usually appear first on the feet and progress upward to belly, hams, ears and sides. In very severe cases, the entire body is covered. The disease is benign. There is little irritation and rubbing is seldom observed. Body temperatures remain normal. Appetites are reduced but the animals continue to eat some. Spontaneous recovery is usually the rule with no change in diet necessary. Recovery can be speeded greatly by turning animals out to pasture. The disease has not been observed in pasture reared animals.

Histopathological studies have shown no apparent disturbances in the tissue of the heart, liver, kidney, spleen and muscle. Skin sections have revealed a tremendous thickening of the cornified epithelium with rows and masses of nuclei scattered throughout. Blood studies have resulted in no clear differences in hemoglobin, red blood cell count, white blood cell count, blood calcium, or blood phosphorous between afflicted animals and normal animals. All indications are that the problem is nutritional in nature, but the fact that it is impossible to produce the disease experimentally casts some doubt on this interpretation. Work should continue on this important aspect of feeding cottonseed meal to swine.

TABLE 1
PRE-PRESSED SOLVENT EXTRACTED
COTTONSEED MEAL AS A PROTEIN SUPPLEMENT FOR PIGS FED IN DRYLOT
(5 pigs per lot in each experiment)

	Experiment I				Experiment II			
	Initial Weight	Daily Gain	Feed per cwt.gain		Initial Weight	Daily Gain	Feed per cwt.gain	
Lot 1 - Corn - SBOM (14% protein)	35.0	1.14	311		46.1	1.62	277	
Lot 2 - Corn - CSM (14% protein)	35.2	1.03	347		46.0	1.02	330	
Lot 3 - Corn - CSM + DSC (14% protein)	35.2	0.81*	384		44.0	0.98	338	
Lot 4 - Corn - CSM + DSC and high level aureomycin (14% protein)	35.0	0.90	371		46.1	1.08	328	
Lot 5 - Corn - CSM (20% protein)	35.0	0.50**	433		45.8	1.32	329	
Lot 6 - Corn - CSM + DSC (20% protein)	35.1	1.00	339		46.0	1.30	363	

* 2 mild cases of dermatitis

** 3 severe cases, 1 mild case

DISCUSSION

Discussion Leader - A. A. Heidebrecht, Paymaster Feeds, Abilene, Texas

- Singleary: In a two year feeding study on pigs of 7-40 pounds initial weight, similar to the one reported by Mr. Ellis, we obtained greater gains - of the order of 1.8-2.0 pounds per day. What were the feed efficiency results you obtained, Mr. Ellis?
- Ellis: The pigs averaged total gains of 200-225 pounds for six months and the feed efficiency was approximately 350 pounds feed consumed per 100 pounds weight gain.
- Sandahl: How much alfalfa was used in the diet you fed, Mr. Ellis?
- Ellis: Five percent.
- Heidebrecht: Is a level of 0.02% free gossypol in the diet safe for swine?
- Barrentine: If the pigs eat the rations well, that level would not be very safe.
- Holley: Some investigators have said that 0.02% free gossypol in the ration is toxic. On the basis of 20% supplemental protein, this level is provided by 0.10% free gossypol in the meal. Usually such meals are toxic. Yet, I have fed some meals to pigs at this level and have lost no pigs. Therefore, I feel that the term "free gossypol" is arbitrary and ill-defined.
- Ward: Mr. Holley, would you say that a meal containing 0.05% free gossypol is non-toxic to swine when added as 20% of the diet yielding a ration containing 0.01% free gossypol?
- Holley: That level would be questionable. However, some of the experimentally produced meals contain 0.03% free gossypol which yields a ration of 0.006% free gossypol. This is apparently a safe level.
- Heidebrecht: Is there a confusion between toxicity and lysine deficiency?
- Lyman: Lysine availability or deficiency should not be involved with toxicity. When lysine was added to the diet for chicks, in which about the same level of protein was used, the toxicity level was not altered.
- Heidebrecht: Dr. Wallace, did you add fishmeal in the swine rations?
- Wallace: No, only corn and soybean meals were used.
- Heidebrecht: The addition of fishmeal and dried whey to diets for chicks seemed to affect growth. Dr. Couch, would you say that this was due to the lysine content of the added materials or to an unidentified growth factor?
- Couch: In the amounts used in our experiments, neither fish meal nor whey would provide enough lysine to be a contributing factor.
- Bryson: Dr. Wallace, is mono-sodium glutamate a source of lysine?
- Wallace: No, DSC, which was used as an additive in our studies, is a commercially available by-product in the production of mono-sodium glutamate. DSC contains 50% protein, thereby having a high lysine content.
- Altschul: I feel that the toxicity problem can be solved, but we must consider whether or not cottonseed meal contains too little lysine to be a competitor with soybean meal for swine feeding.

- Craig: There is sufficient experimental evidence to indicate that a combination of soybean meal and cottonseed meal produces superior weight gains with swine than a diet of either fed alone. A mixture would probably be desirable anyway in order to be able to adjust the formula of the rations with fluctuations in market prices.
- Altschul: Would similar results be obtained with a poor cottonseed meal combined with soybean meal as those obtained with a good cottonseed meal?
- Craig: Most of the cottonseed meals tested for protein quality, when used in combination with soybean meal, have been chemically treated to remove the gossypol and have suffered little heat damage. On swine and chicks, it has been possible to show definite differences, but we have no data available on mixed feeding of rats.
- Colby: The use of a high protein level in the diet has been discussed, but what would happen at a lower level, about 14%, when the soybean meal may not make up the lysine deficiency of the cottonseed meal?
- Wallace: At present we have no answer to that question. We are working with diets containing two levels of solvent-extracted soybean meal, 12% and 18% levels, fed in two lots with and without antibiotics. So far, the pigs are performing better on the 12% ration than on the 18% ration.
- Tucker: We are trying to produce dermatitis with cottonseed meal by adding from 11% to 33% to the ration. We received the best growth at about 22% cottonseed meal supplementation level. These rations were not vitamin supplemented.
- Rusoff: The combined diet containing soybean meal and cottonseed meal gave better growth on calf starters.
- Heidebrecht: Dr. Ellis, do you have any explanation for the lameness in pigs noted in your experiments?
- Ellis: This work with cottonseed meal was the only occasion in recent years where we have seen rickets. I believe that the phytin in the cottonseed meal is responsible and that the pigs are not getting enough phosphorus and vitamin D. With the present recommended levels of vitamin D, calcium, and phosphorus for the diet, I do not believe lameness can be prevented. These recommended levels (of the National Research Council) are adequate, however, when protein supplements other than cottonseed meal are used.

Seminar IV - STATUS OF COTTONSEED MEAL FOR LAYING HENS

Chairman - J. H. Brawner, Southern Cotton Oil Co., New Orleans, La.

COTTONSEED MEAL IN DIETS FOR LAYING HENS

Burt W. Heywang
Southwest Poultry Experiment Station
U. S. Dept. of Agriculture
Glendale, Arizona

My talk is about experiments conducted at the Southwest Poultry Experiment Station to obtain information on the feeding of cottonseed meal to laying chickens. First I want to point out that while the experiments were conducted at our station, they would not have been possible without the help and advice of several people when they were planned or without the excellent co-operation of the Southern Regional Research Laboratory in supplying numerous products and making gossypol determinations. Co-authors of one or more of our publications are A. M. Altschul, H. R. Bird, C. A. Denton, F. H. Thurber, and Miss R. P. Kupperman.

Laying White Leghorns were in the groups in all the experiments. Hydraulic cottonseed meal and soybean meal were of about equal value as sources of protein in diets containing less than 3 percent of animal protein from fish meal or meat scraps. The limiting factors in the feeding of cottonseed meal to layers thus seemed to be the effects of their content of free gossypol and the component that causes pink discolorations in eggs.

Inasmuch as most of the pure gossypol mixed in a diet was inactivated or destroyed after the mixed diet stood for 144 hours, raw decorticated cottonseed and cottonseed meals were included in the diets as sources of free gossypol.

Although they were adversely affected when the dietary levels of free gossypol were 0.016 percent or higher, egg production, weight, hatchability, and diet consumption were not adversely affected when the dietary levels were 0.008 or 0.012 percent. Body weight was not affected when the dietary level was 0.020 percent or less. There was no mortality when the dietary level was 0.025 percent.

When the dietary level of free gossypol was only 0.001 percent, nearly 5 percent of all eggs stored from three days to six months had yolks with discolorations attributable to gossypol. When the dietary level was 0.008 percent, nearly 16 percent of the fresh eggs and nearly 60 percent of all eggs examined had discolored yolks. Both the percent of eggs with discolored yolks and the degree of yolk discoloration increased as the free gossypol content of the diet increased, and both also increased as the length of the storage period increased.

The free gossypol in diets was not inactivated to an appreciable extent by dl-lysine, the amino groups in three other compounds, or by any component of sardine meal.

No fresh eggs or eggs held in a refrigerator for 3, 5, or 7 days had pink albumen. The percent of eggs with pink albumen increased as the length of the storage period increased.

STATUS OF COTTONSEED MEALS FOR LAYING HENS

C. L. Morgan

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The Clemson Agricultural College
South Carolina Experiment Station
Clemson, South Carolina

Results on the use of the various types of cottonseed meal in laying rations are incomplete. Cottonseed meal as the protein supplement for egg production is not as efficient as soybean meal. In some tests weights of the hens have not been maintained as well as with soybean meal. The storage quality of eggs produced on rations containing cottonseed meal may be

affected, although some meals do not affect storage quality of eggs. Both gossypol and cottonseed oil are factors affecting the storage quality of eggs. Hatchability of eggs is reduced when cottonseed meals contain high levels of oil.

Of the various cottonseed meals studied at this station degossypolized solvent extracted meal appears to have the highest nutritive value in laying rations. Pre-press solvent extracted cottonseed meal has not been used in rations for layers.

Summary of laying trial - White Plymouth Rock pullets - 12 weeks -

Supplement	3-15-51	-	5-11-51*	Hen-weight, pounds	
	Prod. %		Hatched %	Start	End
Animal Protein with Soybean meal	42.7		88.4	5.9	5.8
Hydraulic CSM	28.5		83.6	6.2	5.3
Degossypolized solvent CSM	41.3		82.0	6.0	5.8
Screw-press CSM (S 5-4)	28.1		86.3	5.9	5.4
Screw-press CSM (S 5-9/10)	37.2		61.4	6.2	5.5
Soybean Meal	33.6		83.3	5.6	5.7
Animal Protein with Screw-press CSM (S 5-9/10)	35.6		72.7	6.2	5.7

*Unpublished data - confidential

Summary of laying trial - White Plymouth Rock pullets - 12 weeks -

Supplement	1-11-52	-	4-3-52	Hen-weight, pounds	
			Prod. %	Hatched %	
Animal protein (breeder ration)*			51.3	85.8	
Animal protein plus 3.6% Cottonseed oil			43.1	28.6	
Soybean meal*			49.5	75.9	
Degossypolized solvent CSM			49.8	76.8	
Screw-press CSM			48.1	80.9	
Hydraulic CSM			43.4	42.1	
1/2 Hydraulic CSM			41.3	79.2	
1/2 Meat scraps					

*Eggs produced on these rations showed no discoloration after six month's storage.

Summary of laying trial - New Hampshire pullets - 16 weeks
 6-10-52 - 9-29-52*

Supplement	Prod. %	Hen-weight, pounds	
		Start	End
Soybean meal	56.6	5.84	6.03
Degossypolized solvent CSM	52.7	5.63	5.93
Screw-press CSM	49.3	5.74	5.90
Hydraulic CSM	39.8	5.81	5.78
1/2 Soybean meal	44.6	5.69	5.94
1/2 Degossypolized solvent CSM			
1/2 Soybean meal	52.8	5.72	5.86
1/2 Screw-press CSM			
1/2 Soybean meal	52.3	5.73	6.03
1/2 Hydraulic CSM			
Animal protein (breeder ration)	53.0	5.77	5.68
*Unpublished data - confidential			

DISCUSSION

Discussion Leader - K. T. Holley, Georgia Agricultural Experiment Station, Experiment, Georgia

- Anthony: Do other oils or hydrogenated fats affect hatchability when added to the diet of laying hens?
- Morgan: Crude and refined cottonseed oils are the only oils tested which have produced this effect. Other oils, such as peanut and soybean oils, do not affect hatchability; bleaching and hydrogenation reduced the effect of cottonseed oil.
- Holley: Is egg yolk discoloration related to colored gossypol oxidation products formed because of peroxide developments in the fat present in the meal?
- Heywang: I have noted discoloration ranging from light brown to black whether or not the gossypol present was dissolved in oil.
- Holley: Is there not some fat present in the egg yolk? I have observed olive brown color development in vegetable oils high in peroxides when gossypol is dissolved in them. Could not the discoloration of the yolks be due to the development of peroxides in fats which in turn react with the gossypol? Cold storage would delay peroxide development and hence the appearance of the olive brown color.
- Watts: The color reaction in the egg yolk can be reverted by acetic acid and accelerated by ammonia.
- Altschul: Mr. Heywang, could you make some statement now concerning the lowest level of free gossypol at which egg yolk discoloration could be prevented?
- Heywang: I have obtained discoloration with meals of free gossypol content as low as 0.001%. In dealing with discoloration, most people are concerned with egg yolk color, but the development of pink albumens should also be considered. I consider eggs unfit for the market if there is any color present in yolk or albumen, and it is only with meals at levels of free gossypol of below 0.001% that no discoloration would be noted.
- Altschul: The development of egg yolk discoloration due to gossypol in the meal is undoubtedly our most serious problem. We have already established the critical level of free gossypol for chicks and swine, which are feasible levels compared to that (0.001%) evidently necessary for laying hens. To produce meals of a level of 0.005% free gossypol would be almost impossible by the present methods of processing without excessive expense. Our approach to the problem might be to try to reduce the free gossypol in the processed meal. The addition of iron salts apparently does not work. Some work performed here at the Laboratory by Miss Kupperman on the storage of cottonseed meals, cottonseed kernels, and cottonseed-diet mixtures at different temperatures indicates that there is a disappearance of free gossypol as storage time and temperature increase. Perhaps this approach might be adopted as a means of obtaining meals of a free gossypol content low enough for safe feeding to laying hens.

RESULTS OF STORAGE EXPERIMENTS PERFORMED ON COTTONSEED MATERIALS
AND DIET MIXTURES

R. P. Kupperman
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The actual fate of gossypol present in or added to cottonseed materials and to experimental diet mixtures is affected by the:

1. Concentration of gossypol.
2. Conditions of storage.
3. Constituents of the diet materials.
4. Properties of the cottonseed meals and flakes.

Generally, at lower gossypol concentrations in diets and cottonseed material mixtures, the percentage reduction of gossypol was greater than at higher gossypol levels. However, at the higher gossypol levels, the actual loss, reduction or inactivation was greater, as measured by chemical analysis and correlated feeding studies. The preceding was true of cottonseed materials, cottonseed-diet mixtures and gossypol-diet mixtures.

In many cases, with increased storage time and temperature, there was an increased loss of free as well as total gossypol. The fate of gossypol, as free and total, depended on the constituents of the diets and properties of the cottonseed meals and flakes.

Various types of cottonseed materials with and without added gossypol were stored under the same and different conditions of time and temperature.

Together with conditions of storage and gossypol concentration, the properties of the different cottonseed materials themselves affected the fate of gossypol. With some meals, it was possible to account for the loss in free gossypol by analysis as total gossypol. This loss in free gossypol varied from one type cottonseed meal or flake to another, although the conditions of storage and the gossypol concentrations were similar.

In those cases involving the addition of gossypol to cottonseed meals, the added gossypol could usually be accounted for at the time of preparation by analyses for free and total gossypol. Again depending upon the type of cottonseed material, the rate of reduction of free gossypol varied with storage conditions and gossypol concentration. This loss of free gossypol was generally accounted for in the analysis for total gossypol.

In the case of experimental diet mixtures to which gossypol has been added, the conditions of storage and gossypol concentration again affect the fate of gossypol. In all cases the greatest percentage reduction occurred within the first 24-48 hours. Thereafter, the rate of reduction decreased. Rarely could the added gossypol be accounted for at the time of preparation either by chemical analysis for free or total gossypol or by correlated feeding studies.

Further, there appeared to be some unidentified soluble constituent in alfalfa and alfalfa-corn diets that was responsible for the loss, reduction, or destruction of gossypol.

Cavanaugh: In previous reports presented here, there have been discrepancies in the analyses for free gossypol in solvent-extracted or degossypolized meals. In our present discussion, we are speaking of meals of very low free gossypol levels; yet, in the plant when the present analytical methods for determining free gossypol are used, values of 0.02% and 0.04% are considered good checks. We have found that the value obtained varies with the extraction time used in the method. Is this method sufficiently accurate to measure as low as 0.001%, which is the level Mr. Heywang said is maximum to prevent egg yolk discoloration.

Pons: It is true that, by our method, more free gossypol can be extracted with extensions of the extraction period up to about three hours after which the

level of extracted gossypol remains constant. Arbitrarily and for comparison purposes, we limit the extraction period with aqueous acetone to one hour. We have modified the method recently to measure with accuracy at a level of about 0.01%. It is true that the method is not sufficiently accurate to distinguish between trace amounts. Mr. Hoffpauir is the Chairman of a committee of A.O.A.C. working on an analytical method for determining gossypol in mixed feeds.

Holley:

In degossypolized meal, the free gossypol content measured by the present method is very high, yet the meals are not toxic. The dianalinogossypol present in the meal is evidently hydrolyzed during the course of the procedure and is measured as free gossypol. It is obvious that a better definition of free gossypol is needed.

Seminar V - PRE-PRESS SOLVENT EXTRACTED MEALS

Chairman - J. H. Brawner, Southern Cotton Oil Company, New Orleans, Louisiana

IMPROVEMENT IN QUALITY OF COTTONSEED MEALS AND OILS
THROUGH MODIFICATIONS IN PROCESSING PROCEDURES

F. H. Thurber and W. A. Pons, Jr.

Southern Regional Research Laboratory
New Orleans, Louisiana

The proteins of those grains which make up the major portion of mixed feeds for livestock are deficient in some of the essential amino acids, particularly lysine and methionine. For this reason protein concentrates, with a good distribution of essential amino acids, such as cottonseed meal and soybean meal are in great demand. Cottonseed protein contains slightly more methionine and slightly less lysine than soybean meal. The availability of amino acids is perhaps equal in importance to quantity because excessive heat and other factors encountered in processing may modify the proteins and cause the amino acids to be unavailable to the animal's digestive system at the right time to be absorbed for the synthesis of new proteins.

The objective of our research on cottonseed is to improve the quality of cottonseed meal and oil by modifications in processing procedures. Cottonseed is unique among oilseeds in that it contains myriads of minute resinous glands. The chief component of these glands is gossypol, which makes up from 30 to 50% of the weight of the glands and from 0.4% to 1.7% of the weight of the dry meats. Gossypol interferes with the nutrition of chickens and swine and when extracted with the oil it (and other pigments extracted with it) is responsible for color reversion. For these reasons gossypol must be taken into consideration in all of our processing studies.

That processing conditions do affect the nutritive value of the meal and the quality of the oil was demonstrated by preparing meals and oils in a screw press plant under carefully controlled conditions. Meals prepared from low temperature, dry-cooked meats gave much better growth rates when fed to chickens and swine than those from high moisture content meats cooked at high temperatures. Under the latter conditions nearly all of the pigment glands are broken during cooking freeing the gossypol so that it can bind with other components of the meal. During low temperature dry cooking only a few of the pigment glands are broken. Essentially all of those remaining are broken in the screw press, causing appreciable amounts of gossypol to be pressed out with the oil. The resulting meals have a lower total gossypol content than the wet cooked products, a factor which, along with protein availability, may influence their nutritive value. Nearly all of these meals have a very low free gossypol content and can be used in unlimited quantities in the rations of all farm animals.

These experiments have definitely established that superior meals can be produced by the adoption of low temperatures, dry cooking procedures in screw-press mills. Commercial oil mill operators, however, have pointed out some disadvantages to these procedures; such as, decreased press through-put, increased fines, and increased oil color. It seems that a type of conditioning or cooking is needed to retain the processing characteristics of high temperature cooked meats and at the same time, produce meals of high nutritive value.

Although many lines of investigation could be followed in improving cooking and other processing procedures, the following are being given preference:

1. A survey of pre-press solvent extraction oil mills to determine the effect of processing variables on the characteristics of the meal and oil. (This is a cooperative project between oil mills, nutritional laboratories, and this Laboratory.)
2. The preparation of useful and valuable compounds from gossypol to provide an incentive for the removal of gossypol from the seed.
3. Research to obtain more information concerning
 - (a) the reactions and properties of gossypol (a contract project with the University of Tennessee),

- (b) the composition and properties of "bound" gossypol and
- (c) a chemical procedure to adequately measure nutritive value.

4. The preparation of a low gossypol content cottonseed meal (butanone-extracted) to serve as a standard in nutritional investigations.

5. Laboratory investigations on modifications in processing variables to find a means for producing meals equal in nutritive value to the standard meal.

The results of the oil mill survey will be reported in the next presentations, and will be followed by a description of the procedures used in the nutritional evaluation of cottonseed meals and by a report on laboratory research dealing with the effect of processing conditions on nutritive value. The laboratory studies to improve the quality of cottonseed meal and oil will be discussed in the next seminar.

PRE-PRESS SOLVENT EXTRACTED MEALS

The use of pre-pressing followed by solvent extraction is a relatively recent and growing development in the cottonseed processing industry. Current interest in improved processing methods for increasing the nutritive value and extending the utilization of cottonseed meal, without lowering the quality of the oil, prompted a survey of meals and oils currently produced by the pre-press solvent extraction method. Twenty-six complete sets of mill samples of known processing history were obtained through the cooperation of ten mills located in six states. Each set of samples included sufficient meal for evaluation by chick feeding tests conducted by cooperating nutrition laboratories.

As chemical measures of protein denaturation and damage during processing, nitrogen solubility in 0.5N sodium chloride and in 0.02N sodium hydroxide were used. Changes occurring in the distribution of the gossypol present in the seed were followed by means of available methods for free and total gossypol.

The major reduction in nitrogen solubility took place during cooking. Very little reduction occurred in pressing and somewhat more occurred during solvent extraction and meal drying. Calculated as percentage reduction from the values found for the original prepared meals, reduction in nitrogen solubility in 0.02N sodium hydroxide ranged from 13.8 to 26.2% during cooking; 0 to 3.2% during pressing; and 0.9 to 8.7% during solvent extraction and meal drying. Average reduction values were 21% for cooking, 1% for pressing, and 6% for solvent extraction and meal drying. While reduction in nitrogen solubility in 0.5N sodium chloride and in 0.02N sodium hydroxide were roughly parallel, the data suggested that solubility in sodium chloride is apparently a measure of protein denaturation while that in sodium hydroxide is a better measure of protein damage.

One outstanding difference between pre-pressing and normal screw pressing lies in the changes in protein solubility occurring during pressing. Comparable data (in 0.5N sodium chloride) showed that there is an average loss or reduction in nitrogen solubility of 7.9% in pre-pressing as contrasted to reductions as high as 46% encountered in normal high temperature screw presses and reductions of approximately 14% in low temperature screw pressing. The high through-put and shorter residence time of the meals in the press is probably a major factor contributing to the small reductions in nitrogen solubility during prepressing.

Processing conditions during cooking were found to be the major factor influencing the distribution of the gossypol between the meal and oil. Calculated as percentages of the gossypol contained in the prepared meals, the amount bound during cooking varied from 49 to 85%. Where the level of binding was low in cooking (49%), some 24% of the gossypol appeared in the pre-pressed oil. At higher binding levels during cooking, correspondingly less gossypol was removed in the pre-pressed oils. While some gossypol binding occurred during pre-pressing, the values ranging from 0 to 13%, the levels were far below those previously encountered in low temperature screw pressing (29-41%) or high temperature screw pressing (7-46%). A reduction in free gossypol occurred during solvent extraction. However, it was accomplished by removal of the gossypol with the solvent extracted oil rather than by binding to the meal. The pre-pressed oils seemed to be superior to the solvent extracted oils since they showed lower refining losses and lower refined and bleached colors. Upon storage at elevated temperature (100°F.) both types of oils exhibited bleached color reversions with the increase in bleached color being greater for the solvent oils.

In the final meals, free gossypol content seemed to be somewhat constant for a given mill. Four mills produced meals with free gossypol contents ranging from 0.025-0.035% while meals from the other mills averaged close to 0.050%. Considerable variation was found in total gossypol content, the values ranging from 0.7 to 1.3%. Nitrogen solubility in 0.5N sodium chloride ranged from 26 to 46%, while that in 0.02N sodium hydroxide ranged from 65 to 83%.

NUTRITIVE EVALUATION OF PRE-PRESS SOLVENT EXTRACTED COTTONSEED MEALS

Nutritive values of the pre-pressed solvent extracted meals described in the preceding section were determined by chick growth-rate studies and by a chemical index proposed by Dr. C. M. Lyman. To compare results obtained with the same meal at different laboratories a standard, low gossypol content, butanone extracted meal was included in each study and an index value for each meal calculated by dividing the protein efficiency of each meal by that of the standard and assigning a value of 100 to the standard.

Growth rates were determined by the Department of Agriculture, Sacramento, California under the direction of William L. Hunter and by the Poultry Husbandry Department of Louisiana State University. Additional studies are being made at Texas A and M College and at the Beltsville Research Center.

In making growth-rate studies at Louisiana State University, day old chicks are fed a depletion diet for 10 days, two groups (10 chicks each) are then selected to obtain uniformity among groups and are fed the ration to be evaluated for 14 days. The ration contains 12% protein (6% from cottonseed meal and 6% from cornmeal) and the usual vitamin and mineral supplements. At the close of the feeding period feed consumption and increase in weight of chicks are determined, and protein efficiencies and index values are calculated.

At the Sacramento, California station, 20% of protein is used in the diet. The total protein is supplied by cottonseed meal. In other respects, the methods used at the two stations are similar.

In many instances the index values obtained by the two laboratories were in close agreement. When wide variations occur, it is suggested that an average value be reported from at least three different feeding trials.

In Dr. Lyman's procedure a chemical index of nutritive value is obtained by dividing the percent total nitrogen soluble in 0.02N sodium hydroxide by the total gossypol content expressed as percent. If the total gossypol content is less than 0.85%, the value 0.85 is used. In many instances values determined by this procedure were in agreement with those obtained in the feeding studies.

Many of the meals from the pre-press solvent extraction plants had nutritive index values ranging from 75 to 85 (assuming a value of 100 for the standard meal). These values compare favorably with those obtained by low temperature, dry cooking procedures in screw-press plants and provide an additional supply of cottonseed meals suitable for broiler and swine feeding.

DISCUSSION

Discussion Leader - G. C. Cavanaugh, Ranchers Cotton Oil Company, Fresno, Calif.

Cavanaugh: Pre-pressing appears, from these results, to give better meals. However, there might be some difficulty in convincing mill operators to modify the present processing methods. Dr. Thurber, did you obtain any data on the oils?

Thurber: The oils were studied, and it was found that the amount of reversion was a direct function of the amount of gossypol present in the crude oil. Obviously, hydraulic oils which contained small amounts of gossypol do not revert as readily as pre-press solvent extracted and screw press oils, depending on their gossypol content.

Altschul: To summarize, the results obtained in this survey, now almost complete, indicate that the operations of the pre-press solvent extraction plants yield meals of consistently low free gossypol content and high nitrogen solubility. These meals can be more widely utilized than those produced heretofore. The pre-pressed meals are approaching the butanone-extracted meals in nutritive value and differ from screw-press meals only in the extent of damage to the protein inflicted during processing. The outstanding problem now facing mill operators is that of reducing the free gossypol content to a level safe for feeding to laying hens.

**Seminar VI - PRESENT STATUS OF LABORATORY RESEARCH CONCERNING THE EFFECTS OF
PROCESSING ON NUTRITIVE VALUE OF COTTONSEED MEAL**

Chairman - J. H. Brawner, Southern Cotton Oil Company, New Orleans, La.

**RESULTS OF LABORATORY RESEARCH CONCERNING THE EFFECTS
OF PROCESSING ON THE NUTRITIVE VALUE OF COTTONSEED MEAL**

F. H. Thurber, Southern Regional Research Laboratory, New Orleans, La.

A. B. Watts, Poultry Industry Department, Louisiana State University, Baton Rouge, La.

Further research has been initiated in an attempt to produce high quality oils and meals equal to the standard in nutritive value. One of the first experiments dealt with the effect of temperature, moisture, and time on cottonseed flakes (without agitation) in a closed cooker. At temperatures below 230°F. and moistures below 15%, no appreciable changes occurred in the flakes. Evidently, agitation as well as moisture and heat is required to bring about the changes commonly associated with cooking. In the next series of experiments (now in progress) the effect of chemicals, particularly acids and alkalies, on the meats during cooking is being investigated. Since stirring is essential, a Hobart dough mixer is being used as a cooker. It is provided with ribs to increase the agitation, with a thermocouple well for use in temperature measurements and with gas and electric heaters.

Pigment glands and gossypol are still factors which must be considered. When flaked meats contain less than 22% moisture, not all the glands break, even with violent stirring. Neither acids nor alkalies improve the gland breakage. The addition of alkali does improve the color of the oil. For example, when the flaked meats contained 16% water, the meal contained 0.25% free gossypol and the oil 0.33%. In the same type of cook with 1% sodium hydroxide added, the meal contained 0.35% free gossypol and the oil none at all. The oil with the 0.33% free gossypol is subject to color reversion while that containing little or no gossypol is, insofar as we know, not subject to color reversion.

At moisture contents of about 22% or more, flaked meats become plastic when stirred violently and under these conditions all or essentially all of the pigment glands can be broken even at room temperature. In the presence of alkali, gossypol is water soluble and eventually combines with other components of the meal leaving an oil that is essentially free from gossypol. To determine the influence of pH on nutritive value the water level was increased to 35-40% and the pH of the cooks was varied from 4-7.5% by the addition of phosphoric acid or sodium hydroxide. This type of cooking is illustrated by the following typical example:

Flaked cottonseed meats (moisture content 7%) were agitated in the bowl of the Hobart Food Mixer and moistened with water containing sodium hydroxide. The materials were adjusted to yield a moisture content of 35% and a sodium hydroxide content of 0.5% based on the weight of the flakes (3kg. flakes, 15g. NaOH, 1290 ml. H₂O). After 30 minutes agitation at room temperature, essentially all of the pigment glands were broken and heat was then applied for an additional 30 minutes to raise the temperature of the meats to a maximum of 212°F. During the heating period, the oil coalesced to a readily extractable form and the moisture content was lowered to about 7-10%.

The nutritive value of the meal was determined by chick feeding studies of the type described in the preceding presentation. These tests were conducted under the direction of Professor A. B. Watts at the Poultry Husbandry Department of Louisiana State University, and supported in part by the Educational Service of the National Cottonseed Products Association. When approximately 0.5% alkali (NaOH), based on the weight of the meats, was used in the cooker, many of the meats had an index value of about 120 and free gossypol contents below 0.03%. Similar values were obtained when a small amount of phosphoric acid was used, although the free gossypol content was higher. Over 100 meals have been prepared and compared on this scale. Commercial meals range in index from about 40 to 90; many of them rate from about 75 to 85. Crude oils from the alkali cooks are light yellow in color; those from acid cooks are red.

No attempt has been made to determine the modification necessary to adapt these findings

to commercial processing. The purpose thus far has been to establish principles without reference to commercial application.

DISCUSSION

Discussion Leader - T. F. Bridgers, Farmers Cotton Oil Company, Wilson, N. C.

- Bridgers: I am a hydraulic mill operator, and all I can say in the presence of results reported here is that hydraulic-pressed meals still make good fertilizer and catfish bait. Seriously, however, I feel that hydraulic mill operators should strive for better operating conditions. Their greatest concern has always been to achieve the best possible oil extraction, but the gossypol left in the meal has created problems which are of sufficient importance to warrant improvement in processing, particularly cooking conditions.
- Altschul: The butanone-extracted meal was selected as a standard because of its high nitrogen solubility, low free gossypol content, and high nutritional quality. For purpose of comparison of the nutritional values of experimental meals that of the butanone-extracted meal was given an index of 100. Yet, in the experiment just reported, nutritional indices far above 100 were obtained. Would alkali cooking make the meal more digestible?
- Watts: It is possible that the closer the pH of the meal approaches that in the chicks' digestive system, the more digestible the meal becomes. Another possibility may be that a nutritive or growth factor has been produced in the meal during alkali-cooking.
- Richardson: Has anyone tried to improve butanone-extracted meal or considered that some growth factor was removed along with gossypol during butanone extraction?
- Watts: Some of the nutritional results on chicks obtained with meals autoclaved by Miss Jensen indicated a slight improvement in the nutritional value after a five minute autoclaving period. Perhaps toasting the butanone-extracted meal would raise its nutritive value.
- Eagle: How does the butanone-extracted meal compare, by nutritive tests on chicks, with soybean meal?
- Watts: Expressed in terms of grams gained per gram protein consumed, the values for soybean meal were 2.6 - 2.7, for butanone-extracted meal, 1.9 - 2.1, and for commercial casein, 2.8.
- Bridgers: Dr. Thurber, can you explain why a few of the nutritive indices obtained with alkali-cooked meals are low whereas most of the others were 100 or above?
- Thurber: We don't know whether that is due to inconsistencies in chick feeding results or whether the conditions of cooking are sufficiently critical to cause these variations. At any rate, we have prepared or are preparing samples for feeding tests to be made in triplicate by Dr. Watts. These samples will cover the pH range 3.9 to 9.4 at approximate intervals of 1/2 pH unit. We expect to have these results by the middle of January and hope that they will be conclusive with respect to this particular phase of the work.

Seminar VII - THE USE OF FATS IN ANIMAL FEEDS

Chairman - E. J. Cecil, Ranchers Cotton Oil Co., Fresno, Calif.

THE EFFECT OF THE FAT CONTENT OF COTTONSEED MEAL ON ITS NUTRITIVE VALUE

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With an ever increasing adoption of modern processing methods for the production of cottonseed oil and meal, more and more meal is being placed on the market with low oil content. This efficiency in extraction is an accomplishment of which the industry as a whole can certainly be proud. At the same time, it is well to evaluate the practical significance of lowered oil content in the meal in terms of the value of the meal for use in farm rations. As a starting point for our discussion some of the most important functions of fat in farm feeds may be listed. They are as follows:

Functions of Fat

1. Source of essential fatty acids.
2. Source of energy.
3. Improved digestion and absorption of other nutrients.
4. Improved palatability.
5. Improved physical characteristics of feed.

Our problem then is to evaluate the significance of a change of fat content of cottonseed meal in terms of each of these functions.

The important fact to be considered in connection with the first function of fat listed above is that very small amounts of unsaturated fatty acids, those with two or more double bonds, are required by rats and certain other species. In the complete absence of these essential fatty acids the rats fail to grow, the skin becomes rough and unhealthy, necrosis of the tail frequently occurs and the animals finally die. A requirement for essential fatty acids by chicks has been demonstrated by Reiser and it is not improbable that any farm animal would die if completely deprived of these essential fatty acids. The important consideration is that grains, hay, and practically any natural farm feed has sufficient amounts of this type of fatty acid, and a deficiency never occurs in practical diets, but only in synthetic diets completely devoid of fat. There is, therefore, no disadvantage in reducing the fat content of cottonseed meal on the basis of the first function of fat listed above.

In considering item number two, some well established and well known principles should be kept in mind. First, all farm animals can effectively convert carbohydrates to fat for storage and use in their bodies. Second, small amounts of pre-formed fat in the diet can unquestionably be digested and effectively utilized by the animals. Third, the energy content of a pound of fat is 2.25 times greater than that of a pound of carbohydrate.

Now let us look at two sacks of cottonseed meal both of equal protein content; one contains 6% fat and the other 1% fat. In the second case 5 pounds of fat have been extracted by solvent extraction and the difference made up with 5 pounds of hull. Now this difference of 5 pounds of fat is equivalent to 11.25 pounds of digestible carbohydrates. Actually, the conversion of carbohydrate to fat is never quite 100% efficient, and so in reality we would need a little more than 11.25 pounds of carbohydrate to make up the difference between the two sacks of meal.

This difference in actual nutritive value is certainly there, but its practical significance may well be over emphasized. Suppose, for example, that we have cattle on a range with an abundance of dried grass which can supply the carbohydrate or energy needs of the animals. We are then buying cottonseed meal primarily because we need protein. In such a case a change in fat content of the cottonseed meal may be of very little importance.

Item number three concerns improved digestion and absorption of other nutrients. There is abundant experimental evidence which shows that fat in the diet facilitates the utilization of certain other nutrients; among these may be mentioned the fat soluble vitamins and possibly calcium. From a practical standpoint perhaps the effect on the absorption of carotene is of greatest importance. Generally, the experimental feeding trials which demonstrate this beneficial effect of fat have been with rations varying substantially in fat content, for example, from 2.5% fat to 6.5% fat, which is a differential of about 4%. Now let us see what the change in total fat content of a ration is when we substitute cottonseed meal of 1% fat for cottonseed meal containing 6% fat. The differential in the cottonseed meal itself is 5% fat, but, if we are feeding a ration which contains only 10% cottonseed meal, the differential in the total diet is only 0.5%. It is questionable whether a differential of this magnitude can ever cause measurable differences in carotene utilization. A sample calculation showing the fat differential in a beef cattle ration with solvent and hydraulic cottonseed meal is given below.

The Effect of the Fat Content of Cottonseed Meal on the
Total Fat Content of a Beef Cattle Ration

Daily Ration	Fat Contributed to Ration	
	Ration No. I	Ration No. II
	%	%
10 lbs. corn, 3.6% fat	1.26	1.26
16 lbs. hay, 2.4% fat	1.35	1.35
2.5 lbs. cottonseed meal, 6% fat	0.53	—
or		
0.5% fat	—	0.04
Total Fat Content of Ration	3.14	2.65

In special cases where cottonseed meal is fed with rations of unusually low fat content, such as for example cottonseed hulls with only a bare minimum of green hay necessary to supply the necessary carotene, the significance of changes in the fat content of the meal may be somewhat greater than would be anticipated from the sample calculation given above.

The relationship between the oil content of cottonseed meals and the palatability of the products for farm animals is very difficult to measure quantitatively. There are perhaps a few statements on which we might all agree. The first is that if a diet for hogs or chickens is dusty, if it gets into their nostrils, if it gums up in their mouths, or if for any other reason they do not like it, the food consumption will be below normal and the results will not be good. The second fact is that small amounts of oil in a ration does reduce dustiness and hence is of some value for this reason.

Under the heading, improved physical characteristics of feed, we may consider those factors which do not have any direct effect on the animals but which are important from the standpoint of handling the material. Dry powdery meals are more difficult to handle in windy weather; they are more difficult to pellet; losses of material due to sifting through bags are greater and it is more difficult to maintain such meals as homogenous mixtures in mixed feeds. There is no question but that fat improves these qualities.

When we compare high and low fat cottonseed meals in actual feeding trials, the results should reflect a summation of the factors listed above in so far as they apply to the particular type of feeding operation used in the test. Some general conclusions formulated from the results of the work of numerous investigators are given below.

Beef Cattle

A summary of 22 feeding trials comparing hydraulic and solvent extracted cottonseed meals has been prepared by the Educational Service of the National Cottonseed Products Association. In view of the fundamental considerations discussed above, it is not surprising to find that the unweighted averages for the two types of meal were essentially the same. The average daily gain on solvent extracted meal was 1.65 pounds and for the hydraulic meal 1.67 pounds.

This would indicate that the two types of meal are very nearly equal as protein supplements for cattle.

Dairy Cattle

At the New York Agricultural Experiment Station at Cornell, 17 different experiments were conducted to determine the difference in production with dairy rations containing 2.7% fat as compared to rations containing about 6.5% fat. On an average the higher fat rations gave a 4% increase of 4% milk. It must be kept in mind that the fat differential between these two types of rations was about 4%, and that the fat differential which would result from replacing hydraulic cottonseed meal with solvent extracted meal would be more likely about 0.5% for the total ration. It is well to remember that it has been definitely shown that good milk production (probably not quite maximum) can be obtained with rations containing only 2.5% fat.

Swine

In both growth and reproduction tests it has been shown that swine do well on rations containing as little as 0.5% fat. On such rations the feed required to produce a pound of gain is greater. Considering the fat content of all of our common grains, it is evident that any practical ration for swine would contain considerably more than 0.5% fat.

Poultry

At the New Jersey Station it was found that rations containing only 0.1% fat gave almost as good results as a normal ration.

During experiments at the Texas Station on the development of a method for protein quality evaluation by chick feeding tests, tests were also carried out to determine the effect of small amounts of fat in chick rations. In these tests cottonseed meal supplied the only source of protein and the energy source was cerelese (a fat free carbohydrate preparation). The following table shows the growth rate data obtained on the rations without added cottonseed oil as compared to simultaneous tests where the total content of fat was adjusted to 4% by the addition of cottonseed oil. It will be seen that the differences in rate of gain are very small.

The Effect of the Fat Content of the Diet on the Rate of Gain of Chicks

Protein Source	Fat Content of diet	4 week gain	Fat Content of diet	4 week gain
	%	gm.	%	gm.
S.O.M. 1.5% fat	0.7	264	4.0	267
C.S.M.-S-6-76. 0.2% fat	0.1	272	4.0	282
Solvent C.S.M.				
Sample A, 2.0% fat	1.0	277	4.0	298
Sample B, 1.8% fat	0.9	274	4.0	306
Sample C, 1.6% fat	0.8	345	4.0	330

THE USE OF WASTE FATS IN POULTRY FEEDS

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There are approximately 1 1/4 million tons of inedible animal fats produced annually in the United States according to the report of Schweigert et al. (1952). Such fats were used earlier for the manufacture of soap and for other industrial purposes. Competitive materials from other sources have taken the place of animal fats in this regard. Studies have been initiated by the American Meat Institute whereby the use of animal fats in feeds has been investigated. Poultry rations in use today contain relatively low percentages of fat (approximately 4 percent). There are without question a number of advantages which might occur through increasing the fat levels in poultry feeds.

The problem of rancidity is one that certainly received a great deal of attention before tests were actually initiated. The animal fat has been stabilized with a combination of butylated hydroxyanisole, propyl gallate, and citric acid.

Siedler and Schweigert (1953) observed an increase in the weight of the males when 2, 4 or 8 percent fat was added to a practical type broiler ration. In summarizing the results from the latter study, it was indicated that from 2 to 8 percent fat (choice white grease) could be added to chick diets and that the supplements with regard to rate of gain and feed efficiency was at least equal to that of diets not containing the added fat. Results from this study showed that the fat was efficiently utilized when only 2 to 4 percent was added. There was an incomplete utilization of the added fat when the level was increased to 8 percent.

Earlier studies by Reiser and Pearson (1949) showed that hydrogenated vegetable oil could be added to chick rations without deleterious effects on the chicks.

Buckner et al. (1947) showed that 17 percent corn oil could be added to rations for laying hens without deleterious effects.

Couch and co-workers (1949) fed as high as 30 percent soybean oil to laying hens without adverse effects.

Carver and Johnson (1952) reported that the addition of crude corn oil, refined corn oil, soybean oil, wheat germ oil, oleic acid concentrate or linoleic acid concentrate stimulated chick growth as much as 30 percent. These workers postulated that the chick either had a dietary requirement for specific fatty acids or for unknown vitamins present in vegetable oils, or both.

Reed and Couch fed 5 percent stabilized tallow in a broiler test during the summer of 1953. The basal diet in this case contained an adequate fortification with regard to vitamins and unidentified factors. There were 150 birds per group. The addition of the tallow to the basal diet did not increase the weight of the birds, but did improve the feed efficiency and also improved the pigmentation in the shanks and carcasses of the broilers.

Chang, Lyman and Couch have fed 5 and 10 percent cottonseed free fatty acids in standard broiler rations and have found that 5 percent free fatty acids increased growth and improved feed efficiency. Ten percent free fatty acids produced approximately the same results as did the feeding of the 5 percent. It was indicated from these data that the addition of lecithin to the diet with the fatty acids assisted in the utilization of the latter.

In another study the above mentioned workers fed crude cottonseed free fatty acids at a level of 10 percent. A slight reduction in growth occurred as a result of including such fatty acids in a broiler ration at this level although feed efficiency was not adversely affected. It appears quite possible that lower levels of the crude cottonseed fatty acids might be used with satisfactory results. Such an experiment is presently in progress.

Schweigert summarized the advantages of adding animal fats to feeds at the Texas Nutrition Conference (1953) by stating that adding animal fats to feeds controlled dustiness, increased feed utilization, increased palatability, improved the appearance or feel of the ration, decreased the wear on mixing and pelleting machinery, made pelleting easier, and that such a

practice would lead to a more favorable price for fats to be used for this purpose. There is little question but that the addition of the fat to a mixed feed will decrease the dust in the feed and might increase feed utilization and palatability. It certainly would improve the appearance of the ration and it probably would have a favorable effect on the process of pelleting.

There is little question but that the addition of fat to a feed has not resulted in a significant increase in growth but has improved feed efficiency. It seems that the conclusion might be drawn to the effect that the added fat may not be completely utilized by the birds.

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DISCUSSION

Discussion Leader - F. G. Dollear, Southern Regional Research Laboratory, New Orleans, Louisiana

- Dollear: It appears that the addition of fats does not improve the nutritive value of most feeds. However, there may be some optimum levels of fat content for poultry.
- Couch: There seems to be no critical level. Up to 5% has been used successfully, but Dr. Lyman obtained good results with rations containing as low as 0.1-1.0% fat.
- Skinner: The addition of fat improves the palatability of dusty-type rations.
- Lyman: It should be noted that it was necessary to add an anti-oxidant to the fats used in our experiments to prevent development of rancidity. Otherwise, fats in the diet would have become rancid in about 5 days.
- Bryson: Was the addition of anti-oxidants necessary because of the high moisture content

of the fat resulting from incomplete removal of water?

Lyman: Water was used only to neutralize the acidulated soapstock and it was drained off immediately.

Dollear: Has any work been done on the addition of soap to animal diets?

Lyman: No, not up to the present time.

Pons: In adding acidulated soapstocks, did the free gossypol content of the ration increase?

Lyman: No.

Eagle: Acidulated cottonseed soapstocks do not contain gossypol. Rats fed commercially prepared acidulated soapstock at a 22-1/2% level did not suffer from gossypol toxicity.

Lyman: It has been noted that the gossypol content of acidulated soapstocks depends on the length of time they are allowed to remain in the basic state. Prolonged standing reduced the gossypol content.

Sandahl: Dr. Lyman, did you observe any color changes in the diet due to the addition of soapstock?

Lyman: The acidulated soapstocks used in our studies were black in color; therefore, the resulting mixed feeds were discolored.

Skinner: What procedure did you use for mixing, Dr. Couch? We have found that if the fats are homogenized or well blended into the diet, no color changes or rancidity are encountered, even after as long as six months storage.

Couch: The diets were mixed with soapstock in a Hobart Mixer.

Cavanaugh: Would cottonseed hulls be of any potential nutritional value because of the pentosan content?

Rusoff: For ruminants they are of some value.

Lyman: Calorically speaking, 1.0 g. fat is equivalent to 2.25 g. carbohydrate, if both are digestible.

Seminar VIII - METHODS OF MEASURING NUTRITIVE VALUE

Chairman - E. J. Cecil, Ranchers Cotton Oil Co., Fresno, Calif.

INFLUENCE OF AUTOCLAVING AND GOSSYPOL ON THE CHEMICAL AND
NUTRITIVE PROPERTIES OF A SOLVENT EXTRACTED COTTONSEED MEAL

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By using a cottonseed meal of initially high nutritive value it was possible to determine the relative contributions of autoclaving and subsequent "binding" of gossypol to changes in the chemical, physical, and nutritive properties of the meal. The data from chick feeding experiments indicated a progressive decrease in nutritive value as time of autoclaving increased. This reduction was paralleled by similar decreases in nitrogen solubility in 0.02 N sodium hydroxide. There appeared to be no direct relationship between any other property of the meals and their nutritive values.

In other experiments, gossypol was added to the meal prior to autoclaving for 5 and 30 minutes and the resulting samples contained approximately 1% "bound" gossypol. These samples appeared to be identical in chemical, physical, and nutritive properties to control samples autoclaved for the same time.

From the data on these samples, it appears that it may be possible to estimate the nutritive value of a meal by determining its nitrogen solubility in 0.02 N sodium hydroxide.

DETERMINATION OF NUTRITIVE INDEX BY CHICK FEEDING EXPERIMENTS

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The nutritive index of the meals was determined by chick feeding trials conducted as follows: Chicks were standardized by feeding for the first 10 days on a ration composed of yellow corn meal fortified with vitamins and minerals. At the end of this period the chicks were weighed individually. They were then divided into 5-gram weight groups. The allotment of the chicks to each ration was done in such a manner as to place the same number of chicks from each weight group in each ration group. This has been found to reduce the individual variation within each ration group.

Following the standardization period the chicks were placed on the experimental feeding trial. The experimental rations were formulated in such a manner as to supply 12% protein (nitrogen times 6.25). This protein was supplied equally by cornmeal and the cottonseed meal under investigation. Equal parts of starch and sugar were used to make the ration up to 100 parts. Composition of a typical ration is as follows: Yellow cornmeal, 65.0; CSM, 12.8; starch, 9.0; sugar, 9.1; steamed bone meal, 2.0; oyster shell flour, 1.0; salt, 0.5; manganese sulfate, 8.0 gms.; BY-21, 25.0 gms.; B₁₂ supplement (Merck), 25.0 gms.; choline supplement (25%), 20.0 gms.; cod liver oil (2250A-750D), 227.0 gms. The chicks were kept on this diet for a period of two weeks. At the end of this time the chicks were weighed individually and group feed consumption was determined. The total two-week gain was divided by the protein consumed during the period and the result expressed as the gain per gram of protein consumed. The nutritive index for each meal was determined by dividing the average gain per gram of protein for each sample by that obtained for the standard.

A CHEMICAL INDEX FOR THE NUTRITIVE VALUE OF COTTONSEED MEAL

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The nutritional value of the protein in 23 samples of cottonseed meal was evaluated biologically by chick feeding tests using rations in which cottonseed meal constituted the sole source of protein. The samples include meals made by the hydraulic, the solvent, the pre-pressed solvent, and the screw press processes. Wide variations in the nutritional value of the meals made by all processes were found.

Although the free gossypol content of most of the meals was quite low, the total gossypol content of the meals proved to be an important factor in determining the nutritional value of the protein. No consistent relationship was found between the solubility of the protein in 0.5 normal sodium chloride solution and nutritional value, but a very definite relationship was found between the solubility of the protein in 0.2 normal sodium hydroxide and nutritional value.

A chemical index which takes into account both total gossypol content and nitrogen solubility in 0.02 normal sodium hydroxide was devised for the chemical evaluation of protein quality in cottonseed meal. A good correlation was found between the chemical index values and chick growth rates. Without a single exception all meals which showed high nutritional value in the chick feeding tests had a fairly high protein quality indexed value, and all samples which gave low values in the chemical indexed tests gave poor growth rate in the chick feeding test.

The chemical index is calculated as follows:

$$\text{Chemical index} = \frac{\% \text{ nitrogen solubility in } 0.02 \text{ N NaOH}}{\text{Total gossypol expressed as } \%}$$

If the total gossypol content is less than 0.85,
 the figure 0.85 is used instead of the gossypol content.

The values obtained by this calculation are entirely empirical, and it is quite probable that a more exact mathematical relationship between the results of the chick feeding tests and chemical characteristics might be developed. However, it must be recognized that a complicated mathematical formula would probably have little use in the control of mill operations for the production of superior quality cottonseed meal.

It was found that the nutritional value of the meals as determined by chick feeding tests was directly related to the values obtained for lysine availability as determined by rat feeding tests. In order to study the need for additional lysine in diets containing cottonseed meal as the source of protein, chick feeding tests were conducted with and without added lysine. Lysine supplementation of the poor meals resulted in an increased growth rate of chicks amounting to over 100%. Lysine supplementation of the better meals resulted in some improvement in growth rate but the percentage increase was less.

Complete details of the investigations are given in the paper entitled "Evaluation of Protein Quality in Cottonseed Meals by Chick Growth and by a Chemical Index Method", by Carl M. Lyman, Wan Yui Chang and J. R. Couch, *Journal of Nutrition*, Vol. 49, 679 (1953).

THE RAT-REPLETION METHOD FOR ASSAY OF PROTEIN QUALITY OF COTTONSEED MEAL

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The rat-repletion method for assay of protein quality has been employed by C. A. Cabell and I. P. Earle in a collaborative study of cottonseed meals. In this method, adult male rats are depleted on a protein-free diet, then test fed for 10 days so as to consume about 1 gram daily of protein (actually 0.1564 gram of nitrogen). The measure of protein quality is the gain in weight on the test material compared to a given reference protein.

Twelve cottonseed meals produced under different experimental conditions have been assayed by the rat-repletion methods and the results compared with the findings of six other collaborative laboratories using rat growth, chick growth, and microbiological methods. The precision obtained by the rat-repletion method was considered relatively good. In most cases, the results obtained by different methods agreed in differentiating meals cooked at 160°F. as having the highest values and those at 240°F. as lowest, with meals cooked at 180°F. as intermediate. A few extreme variations from the expected behavior are difficult to understand.

NUTRITIVE EVALUATION OF COTTONSEED MEALS
BY THE MICROBIOLOGICAL ASSAY METHOD OF M. J. HORN ET AL.

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Briefly the original method (Horn, M. J., Blum, A. E., Womack, M., and Gersdorff, C.E.F., J. Nutrition, 48, 231 (1952)) consisted of:

- a) Partial digestion of the meal sample by successively subjecting it to the action of pepsin, trypsin, and hog mucosa enzymes for 24 hour periods.
- b) Microbiological assay of the resulting digest for the essential amino acids - arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophane, and valine.

It was found that meals of high nutritive value (rat feeding tests) yielded a large proportion of all the essential acids, while poor meals did not.

A newer modification of the procedure (Horn, M. J., et al. in press) has been proposed as follows:

- a) Digestion with the enzymes as described above.
- b) Use of a single micro-organism and a medium in which all of the essential amino acids are absent. The digest is tested for its ability to induce growth of the micro-organism under these conditions. Hence, the amount of growth obtained is taken as a measure of the ability of the digest to supply all of the essential amino acids.

Under these conditions a good correlation was found between the growth-supporting property of the meal digest and nutritive value of the original meal. This newer method is much shorter than the original procedure and has been applied to various foods, including cottonseed meal. The improvement of the quality of bread protein upon the addition of cottonseed flour was demonstrated.

DISCUSSION

Discussion Leader - E. Eagle, Swift and Company, Chicago, Illinois

Eagle: I would like to dwell a short while on the meaning of the word "significant" as used in reporting the nutritional results here. Is the difference between nutritive indices of 100 and 108, as reported by Miss Jensen for a five minute autoclaved meal, "significant"? If so, it might be well to recommend slight toasting of the meal to improve its nutritional quality. However, the actual "significance" of many of the nutritional data reported here would be elucidated only by a thorough application of biostatistics. Moreover, the five minute autoclaving period produced a reduction in free gossypol content from 0.04% to 0.009%. This should be considered in conjunction with the increase in nutritive value obtained after the same autoclaving period.

At this conference, one confusing point to me is the relative unimportance of free gossypol content as a criterion for nutritive value. At previous conferences it was given more consideration than any other. We have data which shows meals of a free gossypol content as high as 0.4% to be safe and as low as 0.03% to be toxic. However, the shift of interest appears to be away from free gossypol and toward total gossypol.

A more critical characterization of "bound" gossypol appears necessary. Confusion seems to exist between the data of Miss Jensen and that of Dr. Lyman with regard to the effect of "bound" gossypol on nutritive value. Perhaps the data might be less confusing if it could be shown that different types of "bound" gossypol exist, some being more tightly bound than others.

Lyman's data on the influence of total gossypol on nutritive values were impressive, but I would question the use of such high levels of protein to determine nutritive value. At such levels, slight differences in quality between proteins might be masked. There appears to be a definite need to learn more about protein solubility in dilute sodium hydroxide and its relationship to nutritive value.

Lyman: The use of high protein levels in the diet was justified by the agreement in previous nutritional investigations between results from chicks fed low and high concentrations of proteins from the same meal.

Mays: What sodium hydroxide solubility levels and chemical indices would indicate meals of satisfactory nutritive value?

Lyman: From the data I reported, I would suggest that the protein solubility be at least 70% and the chemical index about 80%.

Bruins: Why was cottonseed meal used as the sole source of protein in these experiments when it is usually fed in a mixed diet?

Lyman: It was used alone in order to make its effects more distinguishable.

Watts: It is well known that single sources of protein do not give maximum growth but are employed only to eliminate the confusion resulting from the introduction of another protein source.

Bryson: Is the variation in the properties of the various meals reported by Dr. Lyman due to variations in moisture contents during processing?

Lyman: The meals were of diverse origin, and very little specific processing data is available.

Pominski: Dr. Lyman, do you have nitrogen solubility data on meals with high gossypol content?

Lyman: We have some data of that type, but there does not appear to be a connection

between the solubility and the gossypol content. Dr. Kuiken, when associated with our Laboratory, reported a reduction in lysine availability caused by the binding of gossypol with meal during autoclaving only when the gossypol was added to the meal in oil.

Jensen: Apparently, I did not make it clear that in our experiments, gossypol was added to the meal and autoclaved by the procedure recommended by Dr. Kuiken, that is, in oil and in acetone. Our evaluation of these meals was based on their nutritive value to chicks while Dr. Kuiken's was based on rat feeding experiments and microbiological assays.

Altschul: One of the most important problems facing producers of cottonseed meal now is finding a method for evaluating the meal. We must be able to label a sack of meal with some chemical index of its nutritional quality. The main point of disagreement seems to be on the relationship of "bound" gossypol to nutritive value.

Miss Jensen showed that gossypol "bound" during autoclaving had no effect on nutritive value; while Dr. Lyman found the "bound" gossypol in the meal was related to its nutritive value. Apparently, gossypol binds with the meal in different ways. Miss Castillon has shown that various solvents can remove increasing amounts of gossypol as the length of time of extraction is increased. There is some gossypol, however, that is not extracted under these conditions. Gossypol apparently forms several types of complexes, one of which has been assumed to be a gossypol-lysine complex. We have no definite proof of the presence of such a complex and we cannot have until the complex is isolated from cottonseed meal, perhaps by a partial hydrolysis of the meal.

However, we must remember that the cottonseed industry is not the only group faced with this problem. The soybean meal processors, for example, do not have a chemical index for the protein quality of their product.

DISCUSSION OF ACCOMPLISHMENTS AND RECOMMENDATIONS

Chairman - J. A. Stillwell, Anderson Clayton and Company, Houston, Texas

Stillwell: First, I would like to compliment this group on the excellent work and accomplishments since the last conference in 1951. I believe that areas of differences and of agreements have been elucidated at this meeting and we should take the time now to review our objectives and set up new criteria based on the results reported here. I will leave the technical aspects of the work for others, but from my interpretation of the discussions, the following seem to be the points of agreement:

1. A 50-50 mixture of cottonseed meal and soybean meal can be used in chick broiler rations.
2. Cottonseed meal can be used in swine rations if the free gossypol content is reduced to a certain level.
3. Solvent extracted meal of low fat content can be used in cattle rations.
4. A statement of the relative value of pre-press and straight solvent extracted meals should be possible.

Let us discuss these points before we consider others on which there may be less agreement.

Watts: I would recommend the 50-50 ration of cottonseed and soybean meals for chicks and broilers if the free gossypol content is 0.05% or less.

Heidebrecht: We have been using a ration consisting of 70% soybean meal and 30% cottonseed meal with excellent results. It may be possible to increase the levels of

cottonseed meal by using the improved pre-press solvent extracted meals of today.

Altschul: It seems to me that meals of 0.06% free gossypol content could be used as the sole source of protein in the ration.

Heywang: I don't believe the data reported here would support such a statement.

Craig: Perhaps it could be said that a pre-press solvent extracted meal of 0.04% free gossypol content could be used safely in a 50-50 mixture in the ration.

Lyman: In trying to impose necessary restrictions on the meals for safe feeding practices, would it not seem better to express these restrictions in terms of meal characteristics rather than types of processing?

Altschul: Perhaps we could say that any meal whose nitrogen solubility in sodium hydroxide is 60% or about would be good.

Lyman: Before we make a definite statement, meals of known nitrogen solubility which have been processed in various ways should be tested nutritionally.

Altschul: It seems to me that enough data has been presented to show that cottonseed meal of 0.04% free gossypol content or less and 50% nitrogen solubility or more are equal to or better than soybean meal in nutritional quality.

Eagle: Do we have enough experimental data to make such a definite statement?

Altschul: Maybe this could be a preliminary statement, the experimental proof of which could be collected within the next year.

Lyman: From our experimental data the minimum level of nitrogen solubility for a good meal would have to be 75%, not 50%.

Altschul: On the basis of these discussions then I would like to propose the following resolution:

Results presented thus far indicate that chick and broiler rations containing cottonseed meal and soybean meal in equal proportions on a nitrogen basis are equal to or superior to rations based on either cottonseed meal or soybean meal alone, when the cottonseed meal used has 0.04% or less free gossypol and 75% or more nitrogen solubility in dilute alkali.

Sherman: How will this affect feeds for broilers which are used later as breeders?

Heywang: There should be little effect if laying hens are not permanently on a cottonseed meal diet.

Stillwell: We should now consider formulating another resolution on the second point of agreement.

Wallace: Without considering nutritional effects, it can be definitely said that meals of 0.04% free gossypol or less can be used for swine with no danger of toxic effects.

Altschul: Resolution No. 2 could then be:

Preliminary indications are, insofar as free gossypol is concerned, that cottonseed meals having 0.04% or less free gossypol can be fed in unrestricted proportions in balanced diets for chicks, broilers, and swine.

Ward: We should eliminate the third point of agreement outlined by Mr. Stillwell since no data on cattle feeding was presented at this conference.

- Lyman: Since we have already decided to express our limitations on the use of cottonseed meal in mixed diets in terms of chemical characteristics of the meal we might also eliminate Point No. 4.
- Stillwell: We should consider those subjects on which there has been no general agreement:
1. Egg-yolk discoloration
 2. Effect of lysine on the diet
 3. Lowering of free gossypol content through storage of the meal
 4. Dermatitis in swine
- Altschul: We must decide on a safe level of free gossypol in meals fed to laying hens before we can set up a definite goal to work toward.
- Stillwell: It seems to me that one criterion for this work is the use of laying hens in all experiments.
- Morgan: We should not forget that crude cottonseed oil in the meal can reduce hatchability.
- Lyman: Since Wesson Oil does not have the same effect as crude cottonseed oil perhaps we should try to find out what deleterious constituent is removed from the oil during bleaching and winterizing.
- Craig: Another point which should be considered in future investigations is the effect of the fiber content of various cottonseed meals on their nutritive value.
- Heidebrecht: One of the objections to the use of soybean meal in mixed feeds is the fiber content of 7%. Cottonseed meals have an even higher fiber content, approximately 12 percent. This is a factor to consider.
- Altschul: Can we produce a cottonseed meal equal to or better than a soybean meal when used as the sole source of protein in the diet?
- Eagle: It has already been shown that some cottonseed meals are superior to soybean meals in rat feeding tests.
- Watts: No single protein can be expected to supply all the essential amino acids; combinations must be used for practical rations.
- Lyman: While soybean meal is relatively deficient in methionine and high in lysine, we must remember that cottonseed meal is relatively deficient in lysine but high in methionine.
- Morgan: Cottonseed meal is also high in arginine, an amino acid required by the chick.
- Altschul: There is the possibility of lowering the free gossypol content of rations through storage, but before we can consider the practicality of this procedure, more experimental data must be obtained.
- Craig: Results reported on the incidence of dermatitis indicate that it is never produced when swine are on pasture and that it can be checked by the addition of cobalt and zinc to the rations. However, it has been produced in diets containing peanut meal and shrimp and rice bran meal mixtures. Dermatitis does not seem to be a problem specifically related to cottonseed meal nutrition.
- Altschul: I think we can eliminate the question of lameness brought up by Dr. Ellis since the occurrence of rickets in swine reported by him at this conference seems to be an isolated incident.
- Heidebrecht: Another point which has not been discussed at this conference but which should be considered for future work is the effect of cottonseed meal on reproduction and lactation in swine.

RECOMMENDATIONS

Statement of Policy Regarding Use of Cottonseed Meal in Feedstuffs

- A. Results presented thus far indicate that chick and broiler rations containing cottonseed meal and soybean meal in equal proportions on a nitrogen basis are equal or superior to rations based on either cottonseed meal or soybean alone, when the cottonseed meal used has 0.04% or less of free gossypol and 75% or more of nitrogen solubility in 0.02N NaOH solution.

Nutrition investigators representing industrial, federal, and state organizations are invited to conduct experiments based on this statement using cottonseed meal of known processing history and chemical characteristics and soybean meal of high quality.

- B. Preliminary indications are, insofar as free gossypol is concerned, that cottonseed meals having 0.04% or less of free gossypol can be fed in unrestricted proportion in balanced diets for chicks, broilers, and swine.

Suggestions on Research for Improving the Nutritive Value of Cottonseed Meal

I. Nutritional Investigations

1. Information is needed on comparison of cottonseed meal versus soybean meal as a major source of protein in practical type rations. The following research is suggested:

Chick and hog feeding experiments using broiler type chicks and young weanling pigs should be carried out with selected samples of cottonseed meal of known processing conditions and low free gossypol content (0.04% or less). The cottonseed meal samples should be fed in practical type, nutritionally adequate rations with and without fish meal and should be compared with a ration containing a good quality solvent soybean oil meal. For most of the samples of cottonseed meal, short chick feeding experiments (four to six weeks) should be carried out using protein levels of 20 - 21%. With some of the better quality cottonseed meals full term broiler experiments should be run changing from a 20 - 21% protein starting ration to a finishing ration containing 17 - 18% protein at 7 - 8 weeks. Similar experiments should be conducted with growing turkeys. Rations for starting hogs should contain 16 - 18% protein, and when the hogs reach 100 - 125 lbs. live weight, the rations should be changed to ones containing 14 - 16% protein.

2. Results presented thus far indicate that chick and broiler rations containing cottonseed meal and soybean meal in equal proportions on a nitrogen basis are equal or superior to rations based on either cottonseed meal or soybean meal alone, when the cottonseed meal used has 0.04% or less of free gossypol and 75% or more of nitrogen solubility in 0.02 N sodium hydroxide solution. Further experiments are needed to verify this statement using cottonseed meal of known processing history and chemical characteristics and soybean meal of high quality.
3. Dermatitis in swine, not peculiar to cottonseed meal diets, has interfered with evaluation of cottonseed meal in swine rations. Research is therefore needed to determine the cause of the dermatitis problem and its control so that accurate information will be available on the feeding value of cottonseed meal for swine, unencumbered by dermatitis problems. It is recommended that the assistance of faculties of veterinary medicine be enlisted in an effort to determine the cause of the disease and methods for its control. Information is needed on the symptoms, their extent, their effect on the blood, etc. An accurate description of the morphological skin changes characteristic of this dermatitis is needed. Effort should be made to determine whether an internal metabolic factor or an external agent is the primary cause of trouble. Certain experiments which have suggested the possibility that cobalt and zinc may play a role in dermatitis warrant further study.

4. Information is needed on the effect of feeding cottonseed meal in swine rations on reproduction and lactation. It is recommended that research be conducted to determine the value during reproduction and lactation of cottonseed meal prepared by various processing methods when fed as the only protein supplement and in combination with other protein supplements in swine rations.
5. It is recognized that the procedure whereby a number of different investigators tested the sample groups of cottonseed meal for nutritional value has yielded very worthwhile results, not only for the progress of the cottonseed program, but in improving the reproducibility of nutritional evaluations generally. It is therefore recommended that this procedure be continued and that the meals be collected and distributed from a central agency such as the Southern Regional Research Laboratory.
6. The effect of "bound" gossypol is debatable. With high levels of free gossypol (approximately 0.06% and above) "bound" gossypol has no measurable effect. Many unexplained differences in feeding value of chemically treated cottonseed meal may be due to differences in "bound" gossypol content. It is difficult to separate the effect of unavailable lysine and "bound" gossypol. For these reasons it is suggested that the effect of adding gossypol bound to aniline and gossypol bound to lysine to soybean meal in the diet be determined. Any toxicity would result from the gossypol in these compounds. Preliminary experiments with dianilino-gossypol in soybean meal have resulted in no deleterious effect. The effect of feeding low "free" gossypol pre-press meal containing fairly high "bound" gossypol content should be compared with feeding the same meal in which the gossypol has been sequestered by binding to aniline. This test combined with the first test would show if any reduced feeding value was due to lysine deficiency or "bound" gossypol toxicity. A comparative test should include the addition of all essential amino acids that are known to be required.
7. More information is needed on the effects of high levels of cottonseed meal on calcium and phosphorus metabolism and of phytic acid metabolism in weanling pigs. Experiments at Beltsville have indicated that in the absence of relatively high levels of Vitamin D the weanling pig utilized phytic acid phosphorus more or less poorly with the results that unless sufficient supplementary calcium and phosphorus are added to the diet there occurs a rachitic lameness which varies considerably in severity from litter to litter. On the other hand, over-supplementation of the diet with minerals, while it prevents lameness, appears to favor the incidence of dermatitis.

II. Chemical and Processing Research

1. It is recommended that work be continued and expanded for the development of improved chemical procedures for evaluating protein quality and heat damage in samples of cottonseed meals.
2. It is known that the free gossypol content of cottonseed meal must be 0.01% or less before it can be fed to laying hens in appreciable quantities and not produce egg yolk discoloration. Such a level of free gossypol content has not been obtained by ordinary processing methods or any of the new procedures reported thus far. Research is needed on the development of inexpensive procedures for lowering the free gossypol content of meals containing 0.04% free gossypol to levels of 0.01% free gossypol so that they can be used in diets for laying hens. Methods that should be investigated would include the effects of storage of the meal, chemical treatment, grinding the meal to fine particle size and heat treatments.

Information on the nature of the pigment in the egg yolk may supply clues as to the type of work needed to eliminate it from the meal. There has also been some indication that there are other factors in egg discoloration besides gossypol. These may be related to the residual oil in the cottonseed meal. Additional research to identify such factors in cooperation with nutrition investigators would be very desirable.

3. The cottonseed industry is pioneering a new standard in the feed industry, that of offering products of known free gossypol content and protein quality (presently measured by nitrogen solubility in dilute alkali). Inasmuch as these quality factors are functions of processing conditions, it is important to accelerate the fundamental research on the chemistry of cottonseed processing or gossypol reactions, and on the

production of improved meals by chemical treatment during cooking. The research conducted by Dr. Thurber on alkali-wet cooking should be accelerated and as soon as possible sufficient quantities of meals produced by that method should be made available for feeding tests to chicks, swine, and laying hens.

4. In order to evaluate properly meals from high speed expeller operations, or any meals produced by a radical departure from accepted and known plant operating procedures, a limited study should be made to determine free gossypol content of meal, protein quality, both by chick growth and by chemical index methods, and egg yolk discolorations by the accepted methods of analysis. Preferably the work should be done and meals furnished by industry and analyzed by the Southern Regional Research Laboratory. Whenever possible in this work, representative samples from various sections of the cotton growing belt for any given process should be compared.

Members of the Committee	)	A. M. Altschul, Chairman
on Research, Third Conference on	)	C. M. Lyman
Cottonseed Processing as Related	)	Edw. Eagle
to Nutritive Value of the Meal	)	H. L. Craig
		A. A. Heidebrecht
		W. C. Sherman
		N. R. Ellis
		C. R. Grau
		J. H. Brawner
		G. C. Cavanaugh

SUMMARY OF PROCEEDINGS

A. M. Altschul
Southern Regional Research Laboratory
New Orleans, Louisiana

Over 60 representatives of groups participating in a cooperative program on improving the nutritive value of cottonseed meal through changes in processing conditions attended this conference sponsored jointly by the Southern Regional Research Laboratory and the Educational Service of the National Cottonseed Products Association. Members of the cottonseed processing industry and the feed mixing industry, as well as scientists from commercial, federal, and state research laboratories were present.

The purpose of the conference was to consider the status of progress in obtaining widespread utilization of cottonseed meal through improvements in the processing conditions. Although cottonseed meal had been long considered a valuable protein supplement for beef and dairy cattle, its use in swine and poultry rations had been seriously restricted.

Dr. C. H. Fisher and Mr. A. L. Ward opened the conference with addresses of welcome. Dr. A. M. Altschul of the Southern Laboratory presented a history of the cooperative program which started in December 1948 and was coordinated and encouraged by two previous conferences held in 1950 and 1951. Following this review, seminars were held on the status of the use of cottonseed meals in practical rations for chicks, broilers, swine, and laying hens. General addresses were given by Dr. C. M. Lyman on the effect of fat content of cottonseed meal on its nutritive value and by Dr. J. R. Rouch on the use of waste fats in poultry feeds. Two seminars were presented on the effect of processing conditions on the quality of cottonseed meal and oil. One of these reviewed the status of pre-press solvent extraction methods with respect to the quality of meal and oil, and the second reviewed the laboratory research at the Southern Laboratory on improving the nutritive value of cottonseed meal through fundamental changes in processing conditions. A seminar was devoted to one of the most important problems, namely, that of developing methods of measuring nutritive value.

On the last day of the meeting the entire group met to decide on the general areas of agreement and on the types of future research which would be most beneficial. As a result of this discussion two resolutions were agreed upon as a statement of policy.

Suggestions for future research included work in the following fields: elimination of the problem of egg yolk discoloration associated with cottonseed meal; improving the nutritive value of cottonseed meal when it is the major source of protein; dermatitis in swine; chemical measures for nutritive value; laboratory research on the fundamentals of cottonseed cooking; nature of "bound" gossypol; and effect of cottonseed meal on reproduction and lactation in swine. A committee was appointed to formulate these research suggestions for inclusion in the proceedings. The appointed members were A. M. Altschul, Chairman, C. M. Lyman of Texas A and M College, E. Eagle of Swift and Company, H. L. Craig of Buckeye Cotton Oil Company, A. A. Heidebrecht of Paymaster Feeds, W. C. Sherman of Ralston Purina Company, N. R. Ellis of the Bureau of Animal Industry, C. R. Grau of the University of California, J. H. Brawner of Southern Cotton Oil Company and George C. Cavanaugh of Ranchers Cotton Oil Company.

A Proceedings of this conference will be issued.

PROGRAM

General Chairman - A. M. Altschul, New Orleans, Louisiana

Monday - November 9, 1953

9:30 A.M.

Chairman - J. A. Kime, New Orleans, Louisiana

OPENING REMARKS

C. H. Fisher, Director, Southern Regional Research Laboratory,
New Orleans, Louisiana

A. L. Ward, Director, Educational Service, N.C.P.A., Dallas, Texas

HISTORY OF COOPERATIVE PROGRAM AND REVIEW OF PREVIOUS CONFERENCES

A. M. Altschul, New Orleans, Louisiana

SEMINAR I - Use of Cottonseed Meals in Practical Chick and Broiler Rations

J. R. Couch, College Station, Texas

C. L. Morgan, Clemson, South Carolina

DISCUSSION

Discussion Leader - W. C. Sherman, St. Louis, Missouri

Monday - November 9, 1953

1:00 P.M.

SEMINAR II - Discussion of Pigments and Amino Acids Distribution in
Cottonseed

A. M. Altschul, New Orleans, Louisiana

C. M. Lyman, College Station, Texas

DISCUSSION

SEMINAR III - Status of Cottonseed Meal for Swine

N. R. Ellis, Beltsville, Maryland

H. L. Craig, Cincinnati, Ohio

H. D. Wallace, Gainesville, Florida

DISCUSSION

Discussion Leader - A. A. Heidebrecht, Abilene, Texas

Tuesday - November 10, 1953

9:00 A.M.

Chairman - J. H. Brawner, New Orleans, Louisiana

SEMINAR IV - Status of Cottonseed Meal for Laying Hens

B. W. Heywang, Glendale, Arizona

C. L. Morgan, Clemson, South Carolina

DISCUSSION

Discussion Leader - K. T. Holley, Experiment, Georgia

SEMINAR V - Pre-press Solvent Extracted Meals

F. H. Thurber, New Orleans, Louisiana

W. A. Pons, Jr., New Orleans, Louisiana

DISCUSSION

Discussion Leader - G. C. Cavanaugh, Fresno, California

SEMINAR VI - Present Status of Laboratory Research on Effect of Processing on Nutritive Value of Cottonseed Meal

F. H. Thurber, New Orleans, Louisiana
A. B. Watts, Baton Rouge, Louisiana

DISCUSSION

Discussion Leader - T. F. Bridgers, Wilson, North Carolina

Tuesday - November 10, 1953

2:00 P.M.

Chairman - E. J. Cecil, Fresno, California

SEMINAR VII - Use of Fats in Animal Feeds

C. M. Lyman, College Station, Texas
J. R. Couch, College Station, Texas

DISCUSSION

Discussion Leader - F. G. Dollear, New Orleans, Louisiana

SEMINAR VIII - Methods of Measuring Nutritive Value

E. A. Jensen, New Orleans, Louisiana
A. B. Watts, Baton Rouge, Louisiana
C. M. Lyman, College Station, Texas
N. R. Ellis, Beltsville, Maryland
G. E. Mann, New Orleans, Louisiana

DISCUSSION

Discussion Leader - E. Eagle, Chicago, Illinois

Wednesday, November 11, 1953

9:00 A.M.

DISCUSSION OF ACCOMPLISHMENTS AND RECOMMENDATIONS

Chairman - J. A. Stilwell, Houston, Texas

Meeting of Committee to prepare summary and conclusions of conference. The object of this meeting is to outline the answers to the following questions:

1. Where do we stand on this problem?
2. What type of additional work would be helpful in bringing about wider utilization of cottonseed meal?

Adjournment at Noon

LIST OF ATTENDANCE

Andrews, J. P., Sales Department, Lukenweld, Coatesville, Pennsylvania
 Anthony, W. Brady, Department of Animal Husbandry and Nutrition, Alabama Polytechnic Institute, Auburn, Alabama
 Ball, Charles E., Farm Journal, 5526 Dyer Street, Dallas, Texas
 Barrentine, B. F., Department of Animal Husbandry, Mississippi State College, State College, Mississippi
 Brawner, J. H., Southern Cotton Oil Company, 210 Baronne Street, New Orleans, Louisiana
 Bredeson, D. K., The V. D. Anderson Company, 1935 West 96th Street, Cleveland, Ohio
 Brewster, J. W., Production and Marketing Administration, Washington, D. C.
 Bridgers, T. F., Farmers Cotton Oil Co., Wilson, North Carolina
 Bruins, H. W., Research Laboratories, The Quaker Oats Company, 345 East 25th Street, Chicago 16, Illinois
 Bryson, J. H., Jr., Dothan Oil Mill Company, P. O. Box 458, Dothan, Alabama
 Campbell, R. S., Forest Service, Southern Forest Experiment Station, 2026 St. Charles Avenue, New Orleans, Louisiana
 Cavanaugh, George C., Ranchers Cotton Oil Company, Fresno, California
 Cecil, E. J., Ranchers Cotton Oil Company, Fresno, California
 Colby, R. W., The Dow Chemical Company, Texas Division, Freeport, Texas
 Couch, J. R., Department of Biochemistry and Nutrition, Agricultural and Mechanical College of Texas, College Station, Texas
 Covington, H. E., Mississippi Cottonseed Products Company, Jackson, Mississippi
 Craig, H. L., Technical Division, The Buckeye Cotton Oil Company, M. A. and R. Building, Ivorydale, Cincinnati 17, Ohio
 Davis, Roy B., Plains Cooperative Oil Mill, P. O. Box 509, Lubbock, Texas
 Dillard, E. L., Dothan Oil Mill Company, P. O. Box 458, Dothan, Alabama
 Eagle, Edward, Division of Physiology and Toxicology, Swift and Company, Union Stock Yards, Chicago 9, Illinois
 Ellis, N. R., Animal and Poultry Husbandry Research, Bureau of Animal Industry, Beltsville, Maryland
 Espe, Dwight, Office of Experiment Stations, U.S.D.A., Washington 25, D. C.
 Fincher, H. D., Anderson, Clayton and Company, P. O. Box 2538, Houston 1, Texas
 Gandy, Dalton, Educational Service, National Cottonseed Products Association, Box 286, Tech Station, Ruston, Louisiana
 Harper, Garlon A., Educational Service, National Cottonseed Products Association, 618 Wilson Building, Dallas, Texas
 Heath, John E., Sprout Waldron and Company, Dallas, Texas
 Heidebrecht, A. Allen, Paymaster Feeds, P. O. Box 521, Abilene, Texas
 Heywang, Burt W., Southwest Poultry Experiment Station, U.S.D.A., Route 1, Box 80, Glendale, Arizona
 Holley, K. T., Chemistry Department, Georgia Agricultural Experiment Station, Experiment, Georgia
 Hollowell, E. T., Educational Service, National Cottonseed Products Association, P. O. Box 1036, Atlanta, Georgia
 Holmes, Raiford L., U. S. Tung Oil Laboratory, Bogalusa, Louisiana
 Lewis, Kenneth O., Educational Service, National Cottonseed Products Association, P. O. Box 4470, Tech Station, Lubbock, Texas
 Lyman, C. M., Department of Biochemistry and Nutrition, Texas Agricultural Experiment Station, College Station, Texas
 May, Ralph, The Union Oil Mill, Inc., West Monroe, Louisiana
 Mays, J. R., Jr., Barrow-Agee Laboratories, Inc., Memphis, Tennessee
 McKinney, R. S., U. S. Tung Oil Laboratory, Bogalusa, Louisiana
 Melass, V. H., Agricultural Research, The Dow Chemical Company, Texas Division, Freeport, Texas
 Miller, James W., Feedstuffs, 614 Board of Trade Building, Kansas City 6, Missouri
 Moloney, John F., National Cottonseed Products Association, 19 South Cleveland Street, Memphis 4, Tennessee
 Moore, Walter B., The Cotton Gin and Oil Mill Press, 3116 Commerce Street, Dallas, Texas
 Morgan, C. L., Poultry Husbandry Department, The Clemson Agricultural College, South Carolina Experiment Station, Clemson, South Carolina
 Morgan, Irvin, Jr., Morgan Oil and Refining Company, 600 West Pine Street, Farmville, North Carolina
 Pope, C. W., 8752 Louisiana State University, University Station, Baton Rouge, Louisiana
 Richardson, A. S., The Procter and Gamble Company, P. O. Box 175, Ivorydale, Cincinnati 31, Ohio.

Richardson, D., Department of Animal Husbandry, Kansas State College, Manhattan, Kansas
 Robert, S. A., Jr., Division of Marketing and Transportation Research, U.S.D.A., Bureau of
 Agricultural Economics, Washington 25, D. C.
 Roullard, Fred, Jr., Producers Cotton Oil Company, Box 1832, Fresno, California
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 Louisiana
 Sandahl, Craig W., The Southern Cotton Oil Company, P. O. Box 313, Memphis 1, Tennessee
 Sanders, Carter, Producers Cotton Oil Company, Box 1832, Fresno, California
 Sherman, W. C., Biological Research Laboratories, Ralston Purina Company, Checkerboard Square,
 St. Louis 2, Missouri
 Singletary, C. B., Animal Industry Department, Louisiana State University, University Station,
 Baton Rouge, Louisiana
 Skinner, M. L., Sprout Waldron Company, Muncy, Pennsylvania
 Stillwell, J. A., Anderson, Clayton and Company, P. O. Box 2538, Houston 1, Texas
 Tillman, Allen D., Oklahoma A and M College, Stillwater, Oklahoma
 Tucker, Howard, Department of Animal Husbandry and Nutrition, Alabama Polytechnic Institute,
 Auburn, Alabama
 Wallace, C. W., The Union Oil Mill, Inc., West Monroe, Louisiana
 Wallace, H. D., Department of Animal Husbandry and Nutrition, Agricultural Experiment Station,
 University of Florida, Gainesville, Florida
 Ward, A. L., Educational Service, National Cottonseed Products Association, 618 Wilson Building,
 Dallas, Texas
 Watts, A. B., Poultry Industry Department, Louisiana State University, Baton Rouge, Louisiana
 Woodruff, Ralph, Osceola Products Company, Osceola, Arkansas
 Blalock, Hill, Riverside Oil Mill, Marks, Mississippi

COMPARATIVE DATA^{1/} ON FEED EFFICIENCY OF COTTONSEED MEAL, SOYBEAN MEAL, ETC.

Test Animal	Nutritionist	Meal and/or Ration	Feed/lb. gain	Growth Rate Index
Swine	Wallace	Corn-Soybean meal (Control) (14% Protein)	2.94	
	"	Corn - Prepress Solvent Extracted Cottonseed Meal (14% Protein)	3.38	
	Curtin	Degossypolized Cottonseed Meal in Basal Diet	3.14	
.....				
Chicks	Watts ^{2/}	Standard, Butanone Ex- tracted CSM		100
	Hunter ^{3/}			100
	Watts	Prepress Solvent Extracted		83
	Hunter	CSM - S 16-926		92
	Watts	Prepress Solvent Extracted		110
	Hunter	CSM - S 16-518		83
	Watts	Laboratory cooked cotton- seed meals		120 - 140
	Watts	Soybean Meal		140
	Watts	Casein (Commercial grade)		147
	Morgan	Basal Diet with SEM (21% Protein)	2.45	
	"	Basal Diet with Degossy- polized CSM (21% Protein)	2.45	
	"	Basal Diet with Prepress Solvent Extracted CSM (21% Protein)	2.52	
	"	Basal Diet with SEM (21% Protein)	2.27	
	"	Basal Diet with Degossy- polized CSM (18% Protein)	2.40	
	"	Basal Diet with Prepress Solvent Extracted CSM (18% Protein)	2.64	

^{1/} All data presented at conference.^{2/} Diets contained Corn and CSM, 12% Protein.^{3/} Diets contained CSM - 20% Protein.

ANALYTICAL METHODS

AMERICAN OIL CHEMISTS' SOCIETY OFFICIAL METHOD FOR
DETERMINATION OF FREE GOSSYPOL

This method is used to determine the free (extractable) gossypol and gossypol-like pigments in cottonseed kernels, meals, pigment glands, etc. It is based on a colorimetric measurement of the colored solution formed by a reaction between p-anisidine and the gossypol pigments present in aqueous acetone extracts of cottonseed materials.

PROCEDURE:

Extract a meal sample containing approximately 2.5 mg. of gossypol with 50 ml. of 70% aqueous acetone by constant shaking for 1 hour. Filter, then pipette duplicate 2 ml. aliquots of the filtrate into 25 ml. volumetric flasks. To one aliquot add 3 ml. of the acetic acid solution (1 ml. glacial acetic acid/50 ml. 80% isopropanol); this is the gossypol blank. To the other add 3 ml. of the p-anisidine solution (0.5 g. p-anisidine in above mentioned acetic acid solution); this is the sample. Heat the sample and a reagent blank (2 ml. aqueous acetone and 3 ml. p-anisidine solution) at 60°C., with flask loosely stoppered for one-half hour, then cool to room temperature. Dilute the sample, reagent blank, gossypol blank, and solvent blank (2 ml. aqueous acetone and 3 ml. acetic acid solution) to 25 ml. with 80% isopropanol.

With an Evelyn colorimeter (470 filter), determine percent transmission of the sample, T_1 , using the reagent blank to set the instrument at 100% transmission. Determine percent transmission of the gossypol blank, T_2 , using the solvent blank to set the instrument at 100% transmission.

Calculate $\log T_1/T_2 \times 100$ and using this value read off the mg. of gossypol in the sample aliquot, A, from a previously prepared standard curve.

$$\% \text{ Gossypol (Free)} = \frac{A \times 25}{\text{Weight of sample (grams)} \times 1000} \times 100$$

REFERENCES:

Pons, W. A., Jr. and Guthrie, J. D., J. Am. Oil Chem. Soc., 26, 671 - 676 (1949).

American Oil Chemists' Society, Official and Tentative Methods, 2nd Edition (Rev. to 1952).

A MODIFICATION OF THE p-ANISIDINE METHOD FOR THE
DETERMINATION OF FREE GOSSYPOL

W. J. Miller
The Buckeye Cotton Oil Company
Memphis, Tennessee

The free gossypol content of cottonseed meal can be reduced by treating the meal with aniline. This process is being used on a commercial scale at the Buckeye Cotton Oil Company in Memphis, Tennessee. Comparison of feeding tests on these meals with the free gossypol measurements made by the Official Method of the American Oil Chemists' Society indicated that this method gave abnormally high results. This was attributed to a liberation of some gossypol from the dianilinogossypol complex and subsequent reaction with p-anisidine, increasing the final color of the sample. It was found that this error could be eliminated by using aniline in the developing solution rather than p-anisidine.

The most reproducible and accurate results were obtained when the acetone solution of gossypol was reacted with aniline at 100°C. for 15 minutes. This method was found to be applicable to all types of cottonseed meal produced today.

DETERMINATION OF NITROGEN SOLUBILITY IN DILUTE SODIUM HYDROXIDE

"One-gram samples of cottonseed meal ground to pass a 60-mesh screen were placed in 300-ml. Erlenmeyer flasks with 4 glass beads. One hundred milliliters of 0.02 normal sodium hydroxide were added and the flasks were shaken rapidly on a mechanical shaker in an incubator at 37°C. for one hour. The material was then centrifuged at 3,000 r.p.m. for 5 minutes. It was filtered through coarse filter paper if floating particles made this necessary.

"Fifty milliliters of the supernatant liquid were pipetted into a Kjeldahl flask. Two milliliters of concentrated sulfuric acid were added and the material was evaporated almost to dryness on the digestion rack, after which the Kjeldahl determination for total nitrogen was carried out as usual. Nitrogen solubility is expressed as percent of the total nitrogen which is soluble in 0.02N sodium hydroxide."

C. M. Lyman, W. Y. Chang and J. R. Couch
J. NUTRITION, 49(4), pp. 679-690, (1953).

The use of trade names in this publication is for identification and implies no endorsement by the Department of Agriculture of the product over similar products not mentioned.

